1957

# Studies on the passively acquired antibodies in the baby pig in relation to early weaning

Vaughn Curtis Speer
*Iowa State College*

# STUDIES ON THE PASSIVELY ACQUIRED ANTIBODIES IN THE BABY PIG IN RELATION TO EARLY WEANING

by

Vaughn Curtis Speer

A Dissertation Submitted to the
Graduate Faculty in Partial Fulfillment of
The Requirements for the Degree of
DOCTOR OF PHILOSOPHY

Major Subject: Animal Nutrition

Approved:

Signature was redacted for privacy.

In Charge of Major Work

Signature was redacted for privacy.

Head of Major Department

Signature was redacted for privacy.

Dean of Graduate College

Iowa State College
1957

TABLE OF CONTENTS

# INTRODUCTION

The transmission of passive immunity from the mother to the young was first demonstrated in the mouse by Ehrlich (1892). He showed that maternal antibodies were transmitted to the young *in utero* and after birth by way of the mammary secretions. These two methods of transfer vary between species. It is known that antibody production in the newborn is negligible or nil for some time after birth, and so the antibodies acquired by the newborn play an extremely important role in the survival of the young.

The difficulties encountered in trying to raise domestic animals which have not received colostrum or first milk may be cited as evidence of the importance of passive immunity for defense against disease.

In ungulates, colostrum antibodies obtained with the first feedings rapidly pass from the intestine into the blood circulation. This absorptive capacity of the gut has been assumed to be transitory and assumed to be lost within a short time after birth. In his work with baby pigs, Nelson (1953) was able to raise one group of pigs on liquid sow milk substitutes in a "non-germ free" room without colostrum, but further attempts were unsuccessful because of disease problems. When the baby pigs were left on the sow for two or three days, they could be raised successfully in concrete-floored pens on synthetic milk rations. Following this work, baby pigs one week of age were routinely weaned and placed on dry diets with performances equal to or better

than the performance of baby pigs allowed to nurse the sow for the standard eight week lactation period (Speer et al., 1954). Recently Petersen and Campbell (1955) reported antibody absorption via the intestinal tract in 5-month-old calves, adult pigs and adult humans. This apparent contradiction of early work raises some doubts as to the advisability of early weaning of baby pigs even though they may have nursed the sow long enough to insure colostral intake.

The purpose of this research was to investigate the transfer and supplementary value of passively acquired immunity in baby pigs. Three approaches were made on this problem: (1) the comparison of the blood serum proteins in early weaned pigs as compared to pigs nursing sows; (2) the length of time after birth that the baby pig is able to absorb a specific antibody; and (3) the value of supplementary antibody containing materials on the growth of early weaned pigs.

# LITERATURE REVIEW

In the review of the literature several aspects were considered to fully explain the results of the problem studied. It is recognized that there is a difference between species, but because of possible similarities, the inclusion of these reports seemed justified.

## Route of Antibody Absorption in the Young of Domestic Animals, Laboratory Animals, and the Human

### Swine

Though it may have been suspected that the baby pig obtained antibodies passively via the colostrum, the earliest mention of this in the literature was brought out in the discussion of a paper by Hayes (1921). In an experiment dealing with swine abortion, the offspring of sows infected with Bacterium abortus were born without agglutinins, but agglutinins were present in the offspring soon after nursing. The colostrum was also rich in agglutinins and complement-fixing antibodies. Nelson (1932) used vaccinia virus as an immunizing agent to study the route of maternal transmission of antibodies in swine. Young sows were vaccinated immediately prior to and shortly after breeding. At parturition part of the baby pigs were allowed to nurse their dams, and part were fed a mixture of dried cow's milk and normal swine serum. On the seventh day after farrowing, the pigs were vaccinated and kept under observation for 10 days or more. The pigs suckled by immune dams

showed no vaccinia reaction. The hand-fed pigs all responded to the presence of the virus with the formation of typical skin vesicles. Young pigs nursing non-immune dams also showed skin vesicles when vaccinated. Nelson (1932) concluded that the porcine placenta was impermeable to any appreciable amount of protective substances, and that the function of immunity transfer was assumed by the colostrum. Earle (1935) studied the proteins of the blood sera of baby pigs. Using the sodium sulfate precipitation method of Howe (1921), Earle (1935) arrived at the same conclusions with baby pigs as Howe (1921) did with the newborn calf. The baby pig was born with little or no euglobulin, but after receiving colostrum the blood did contain a large amount of euglobulin.

Though Deutsch (1947) did not study the transfer of immunity from dam to offspring, electrophoretic analyses using the Tiselius apparatus, revealed a slow moving component in sow colostrum whey fraction accounting for the largest share of protein present. This fraction, which is associated with the immune globulins and which has a mobility nearly the same as gamma globulin, decreased rapidly a few days post-partum. Foster et al. (1951) noted similar changes. In one of their samples obtained immediately post-partum, the slow moving component accounted for 70 percent of the whey protein. Their study also included data on baby pigs. The gamma globulin content in the pigs' plasma before nursing was as low as 2.7 percent, but rose to as high as 40 percent at 24 hours post-partum. When baby pigs were removed from their dams at birth, Barrick et al. (1954) were unable to detect any gamma

globulin in the blood sera 24 hours later. The blood sera of baby pigs nursing their dams contained 41 to 46 percent gamma globulin 24 hours after farrowing.

Several researches concerned with specific disease entities have shown that little if any placental transfer of immune bodies takes place in utero in swine. Young and Underdahl (1949) detected no hemagglutination inhibitor antibodies against swine influenza in 37 baby pigs at birth. Thirteen pigs did show low titers. The authors felt that these low titers were due to naturally-occurring inhibiting substances, and concluded that placental transfer of swine influenza virus antibodies did not pass the placental barrier in swine. Significant titers were detected in the baby pigs' blood sera within 30 minutes after they had begun to suckle. Later, Young and Underdahl (1950) tested for both hemagglutination inhibitor and neutralizing antibodies against swine influenza virus in baby pigs farrowed by sows challenged with the disease, and arrived at the same conclusion. Both hemagglutination inhibitors and neutralizing antibodies were present in the colostrum of the dams as shown by the high titers. In his studies of swine brucellosis, Hoerlein (1952) was unable to isolate Brucella melitensis from any of the baby pigs farrowed by infected sows. The baby pigs showed no agglutination titer at birth, but a maximum titer was reached in a matter of hours after receiving colostrum from their dams.

It is possible to raise baby pigs that have not received colostrum, but for any degree of success it has been necessary to rear

them in isolation units under germ-free conditions (Young and Underdahl, 1953; Young et al., 1955; Hoerlein et al., 1956; Shuman et al., 1956; Whitehair and Thompson, 1956).

Bovine

The published evidence on the bovine favors the view that no placental transfer of antibodies occurs and that the colostrum is all-important in respect to the transference of passive immunity from the cow to the calf. In biochemical studies on the blood sera of calves, Howe (1921) concluded that the blood of the newborn calf did not contain euglobin or pseudoglobin I. After the calf had nursed, the blood did contain large amounts of these two protein fractions. Later, Howe (1922) used the same anhydrous sodium sulfate precipitation method in an investigation of the proteins of colostrum. His data indicated that the euglobin of colostrum and blood serum were the same. It was not until the electrophoretic technique was developed that slight differences in the two euglobins were found. Little and Orcutt (1922), Orcutt and Howe (1922), and McAlpine and Rettger (1925) used an antigen-antibody immunological technique to demonstrate the lack of antibodies against Bacillus abortus in the sera of the newborn calf, even though the bovine dam had a high blood serum titer. Tests on the blood from calves a few days after birth showed that a high titer against this organism was present. Using the sulfate precipitation method, Orcutt and Howe (1922) were able to associate Bacillus abortus agglutinins with the globulins of colostrum and with the simultaneous appearance of the globulins in

the blood of the newborn calf upon receiving colostrum. McDiarmid (1946) worked with Br. abortus and Henning (1953) worked with S. dublin and both arrived at the same conclusion as earlier workers that the calf is born without antibodies, but acquires them upon the ingestion of colostrum. The protective substances contained in the non-fatty fraction of colostrum and the changes in the blood sera of calves at the first feeding has been described in several papers by Aschaffenburg (1949) and Aschaffenburg et al. (1949a, 1949b, 1951, 1952). Mason et al. (1930) immunized a cow with diphtheria toxin. The calf was born without diphtheria antitoxin, but a small amount was detected after nursing. Several papers by San Clemente et al. (1943), Smith (1946, 1948), Smith and Holm (1948), Smith and Coy (1946), Smith et al. (1946), Smith and Green (1947), Hansen and Phillips (1947, 1949), and Polson (1952), in which the Tiselius electrophoretic apparatus was used to study the colostral and blood serum proteins, confirmed the sulfate precipitation studies of Howe (1921), but expanded the work to show that adult gamma globulin, immune lactoglobulin of colostrum, and the passively acquired gamma globulin fraction in the newborn did differ slightly in mobility, amino acid content, carbohydrate content, and absorption spectra. The significance of colostrum to the newborn calf was demonstrated by Smith and Little (1922) and Smith and Orcutt (1925) in their attempt to raise calves without colostrum. Twelve calves receiving no colostrum died. Bacillus coli was isolated as the causative agent in most of the calves at death. Though this organism was not ordinarily considered virulent, colostrum apparently contained protective qualities that enabled the colostrum-fed

calves to ward off invasion by this organism. Results typical of that observed by Smith and Little (1922) have been observed by a number of laboratories, and the same apparently avirulent organism has been incriminated as the agent that causes difficulties in raising calves without colostrum (Smith and Little, 1930; Aschaffenburg et al., 1951, 1952; Briggs, 1951; Briggs et al., 1951; Roberts et al., 1954; Roy et al., 1955a, 1955b).

Sheep and goat

Famulener (1912) observed that the placenta plays a very small role in the transfer of antibodies in the goat. He immunized goats with sheep erythrocytes during the latter part of gestation. He decided that the principle route of hemolysin transfer was through the colostrum, and thence through the gastro-intestinal tract and absorbed into the blood stream of young kids. This was similar to the results published by Reymann (1920) on agglutinins, Mason et al. (1930) on lamb dysentery antitoxin, Oxer (1936) on lamb enterotoxemia antibodies, and by Filmer and McClure (1951) in their work on the absorption of anti-nematode antibodies in the lamb, as measured by a hemolytic complement fixation test.

Using the "salting-out" precipitation method, Earle (1935) determined that the lamb and kid were born with little or no euglobin and pseudoglobin I, but that following colostral intake, these fractions made their appearance in the blood serum. Electrophoretic analyses later confirmed these observations (Charlwood and Thomson, 1948; Hansen and Phillips, 1949; McCarthy and McDougall, 1949, 1953).

Horse

Mason et al. (1930) found no diphtheria antitoxin in the blood of a foal at birth, though the dam was immunized against diphtheria. The antitoxin was present in the foal's blood after nursing. Earle (1935) showed that the blood picture in the newborn foal was similar to the calf. Polson (1943), employing the electrophoretic technique, did not detect gamma globulin in the newborn foal, but upon nursing, gamma globulin appeared in the blood serum. Howard and Cronin (1955) reported a field case of a mare with a known history of anti-erythrocyte antibody production in her colostrum and milk. Her third foal was nursed on a foster mare until 13 days of age. Twenty-four hours after being placed with its own dam, the colt's erythrocytes were found to be weakly sensitized, but no clinical signs of hemolytic disease appeared as the red cell count and hemoglobin remained within normal limits. Other reports on this problem have been reviewed by Brambell et al. (1953). Contrary to Howard and Cronin (1955), Lemétayer et al. (1946) found very small, but measurable quantities of antitoxin in the blood of newborn foals before they had suckled mares with high titers of tetanus or diphtheria antitoxin in their blood.

Rat and mouse

Ehrlich (1892) has been credited with the observation that maternal antibodies in mice were transmitted to the young in utero and via the mammary secretions after birth. He also showed that highly immunized male mice crossed with normal females failed to transmit resistance to the offspring. In his work with the protozoan parasite Trypanosoma

*lewisi*, Culbertson (1938) studied the route of acquired immunity in the young rat. A definite temporary immunity to the live organism was produced in the young of infected mothers. Subsequent litters were also immune, but the power to confer resistance did diminish with time. By exchanging litters, it was possible to determine whether the passage of immunity took place *in utero* or through the milk. Challenge experiments with the protozoan organism revealed that some protection was passively conferred *in utero*, but better protection was passively acquired through nursing the colostrum milk. Halliday (1955) used an antigen-antibody agglutiniation test to determine antibody absorption in young rats. Immune sera was prepared in the rat, mouse, rabbit, cow, and a fowl and was administered orally to young rats. Agglutinins produced in the mouse were absorbed nearly as well as those produced by the rat. The antibodies produced by the rabbit were absorbed more slowly and to a lesser degree, while agglutinins produced by the cow or in the fowl did not appear to be absorbed at all by the young rats. Halliday's later work (1956) confirmed again that the young rat is capable of absorbing antibodies from the intestinal tract after ingesting colostrum or immune serum. Brambell and Halliday (1956) also worked on the route of antibody transfer from the mother rat to the fetus. Immune *Salmonella pullorum* serum administered via the mouth of the rat fetus was rapidly absorbed from the gut and appeared in the fetal blood circulation. Immune serum was also absorbed from the yolk-sac splanchnopleur. Antibodies were found in the circulation when both of these routes were ligated. They concluded the antibodies were probably absorbed from the cavities in the

entodermal sinuses of Duval, or possibly by direct passage from maternal circulation across the placenta. Bangham and Terry (1957) iodinated homologous and heterologous serum proteins with $I^{131}$. After oral dosage of the labelled serum proteins, serum samples from young rats were separated electrophoretically on a cellulose column. The radioactivity curves of the sera of young rats had well defined peaks corresponding to the gamma globulin fraction of the adult homologous sera. Heterologous labelled sera from rabbits and monkeys were absorbed to a much lesser degree than the homologous sera.

Rabbits and guinea pigs

Brambell et al. (1951, 1954) have summarized the reports on maternal transmission of antibodies in rabbits and guinea pigs. There was general agreement that the serum titer of the newborn before suckling was equal to that of the maternal serum. In their experimental work and that of Batty et al. (1954) from the same laboratory, rabbits injected with antigens or homologous and heterologous antisera during pregnancy gave birth to young which showed high agglutination titers at birth. Similar results were obtained by Cohen (1950).

Dogs

Newborn puppies were able to absorb diphtheria antitoxin sera produced in horses and administered by mouth (Mason et al., 1930). When the antitoxin was administered to a bitch four days prior to parturition, the pups were born with no detectable antitoxin, but it was detectable shortly after nursing.

Man

McGirr (1947) points out that colostrum in the human contains few antibodies, and apparently plays no significant role in the transmission of early passive immunity to the infant. The summary by Brambell et al. (1951) also indicates that passive immunity takes place almost entirely before birth.

Summary

Attempts to classify the route of passive immunity transfer have been made by Schneider and Szathmáry (1938, 1939) and more recently by McGirr (1947) on the basis of placental type. Brambell et al. (1951), as a result of their studies with the rabbit, concluded there was insufficient evidence to support or refute the postulate of preferential antibody transmission via the placenta over other embryonic membranes. Nevertheless, such a classification warrants attention, because the importance of the in utero environment or colostrum as the source of antibodies appears to be correlated with the complexity of the placenta. Table 1 summarizes the information on different animals with respect to placental type and passively acquired antibodies.

## Age As a Factor in Antibody Absorption from the Gastrointestinal Tract

Culbertson (1938), in his study on the transmission of immunity against the parasite Trypanosoma lewisi, transferred 15-day-old rats from non-immune mothers to immune mother rats. An injection of one

Table 1. Classification of animal species, placental types, the route of antibody absorption, and the time absorption occurs after birth[a]

| Animal species | Placental type | Antibody transmission | | Time from birth antibody absorption occurs from intestinal tract[b] |
|---|---|---|---|---|
| | | In utero | Colostral | |
| Man | Hemochorial | ++ | ± | A few days |
| Mouse | Hemochorial | + | ++ | Approx. 20 days |
| Rat | Hemochorial | + | ++ | Approx. 20 days |
| Guinea pig | Hemochorial | ++ | - | Approx. 10 days |
| Rabbit, early fetus | Hemochorial | ++ | - | Approx. 10 days |
| Rabbit, late fetus | Hemoendothelial | | | |
| Dog | Endotheliochorial | ± | ++ | Approx.10-12 days |
| Sheep | Syndesmochorial | - | ++ | Less than 4 days |
| Goat | Syndesmochorial | - | ++ | Less than 4 days |
| Cow | Syndesmochorial | - | ++ | Approx. 1 day |
| Pig | Epitheliochorial | - | ++ | No data available |
| Horse | Epitheliochorial | - | ++ | No data available |

[a]Adapted from McGirr (1947).

[b]Includes data reported through approximately 1946.

million T. lewisi organisms was then given to the transferred young one day after transfer. These young rats proved to be immune, and it was concluded that the intestinal tract of the nursing rat was permeable to protective antibodies ingested from immune milk for as long as 15 days after birth. In another experiment Culbertson (1939a) orally dosed rats that were 10, 15, 20, 25, 40 and 60 days old with antiserum against T. lewisi once daily for 5 days. They were infected with one million T. lewisi parasites intraperitoneally on the first day the antiserum treatment began. Good protection against this parasite occurred in rats 20 days old or less. Brambell et al. (1954) and Halliday (1955) studied the administration of immune rat serum and heterologous immune sera produced by mice, rabbits, cows, and fowl. Young rats up to 18 to 20 days of age readily absorbed antibodies produced by rats, mice, and rabbits into the circulation, while agglutinins prepared in cows or fowl were not absorbed. In a later experiment, Halliday (1956) fostered young rats from non-immune mothers to immune mothers. He also fed young rats immune sera by stomach tube to determine at what age the ability to absorb antibodies from the intestinal tract was lost. The young rats ceased to absorb antibodies against Salmonella pullorum from milk or immune serum from the gut at 20 days of age. Antibodies were still detectable in the milk of immune mother rats after the young had reached this age.

McCarthy and McDougall (1949, 1953) combined three methods to study the changes in serum proteins of suckling lambs that received colostrum after various periods of delay from birth. The methods included electro-

phoretic and precipitation examinations of the serum proteins, and an antigen-antibody agglutination test employing Salmonella typhosa as the antigen. The ewes were challenged with the organism 21 days prior to their expected lambing date. Seven lambs were allowed to ingest colostrum at birth, and one lamb each was allowed colostrum after delays of 6, 12, 24, 29 and 48 hours and after 3 and 9 days. Four lambs were deprived of colostrum. Both the "salting-out" and electrophoretic analyses showed a marked increase in the globulin content of the lamb sera after the ingestion of colostrum at birth with a smaller increase after colostrum ingestion was delayed for 24, 29, and 48 hours. No immediate increase was evident when the colostrum was delayed more than 48 hours or not given at all. Agglutination tests clearly showed absorption of antibodies in the lambs receiving colostrum up to and including 29 hours after birth, but not after 48 hours or more delay. Agglutination titers of animals showing absorption rose from negligible values at birth to a positive test between 1:125 to 1:5000 dilutions after ingestion of colostrum.

McAlpine and Rettger (1925) restrained three calves from nursing for the first 24 hours after birth. Their dams were reactors to Bacterium abortus. The calves were then fed milk which contained reacting antibodies. Later tests on the calves' sera were negative for both agglutination and fixation tests. The authors interpreted these data as a necessity of colostrum for passage of reacting bodies from the alimentary tract to the blood stream of young calves. It was more likely these 24-hour-old calves had lost their ability to absorb intact anti-

bodies. Smith (1946) fed immune serum to two calves, 2 and 18 days old, respectively, and observed no increase in serum agglutinating antibodies. In their electrophoretic studies Hansen and Phillips (1947) fed calves that had not previously nursed specified amounts of colostrum. In no case was there an increase in the gamma globulin fraction in the blood sera of the calves when the calves were more than 24 hours of age. Henning (1953) collected Salmonella dublin antiserum from hyperimmunized cows, and administered this material to calves before they were allowed to nurse their dam. The calves were able to absorb a large portion of the agglutinins from the immune serum. Six calves were then allowed to nurse their dams. The immune serum was administered by mouth to three calves 24 hours after birth, two calves 36 hours after birth, and one calf 48 hours after birth. The author attributed the lack of absorption of antiserum agglutinins in these calves to the intake of colostrum after birth. This was another instance that undoubtedly should be interpreted as a loss of ability to absorb antibodies due to an age factor.

It has been reported that foals 5 days of age can safely nurse a mare whose colostrum contains antibodies capable of reacting with the foal's erythrocytes and causing a fatal hemolytic jaundice or hemolytic icterus. It was also possible to milk mares at hourly intervals for periods of 24 to 36 hours, and then allow the foals to nurse without the development of hemolytic icterus. This procedure reduced the titer of the mare's colostrum and presumably reduced the amount of anti-erythrocyte hemolysin absorption by the foal (Bruner et al., 1950). Nine foals were also fed mare serum having a titer of 1:10,000 against Salmonella

abortivoequina. Each foal received 1000 cc. of serum in a single feeding through a stomach tube. Three of the foals were less than 12 hours old, two were between 12 and 24 hours, three were between 24 and 36, and one was 72 hours old at the time of feeding. Agglutinins were absorbed by the foals less than 24 hours of age. The foals that had attained an age of 24 to 36 hours or more did not show any agglutinins for S. abortivoequina at a titer of 1:10.

Bruner et al. (1949) sensitized several gilts to the erythrocytes of the boar to which they were bred. The majority of the pigs farrowed had the same type of red cells as the boar. Although they were born with normal red cell counts, the pigs were all dead within 12 to 42 hours. Post mortem examination revealed an anemic condition, and in some of the pigs it was difficult to locate any red cells. Samples of the colostrum and the milk from the sows contained hemagglutinins against the red cells of the baby pigs. Normal 2-day-old pigs which were allowed to nurse a sensitized sow did not absorb hemagglutinins even though the sow's milk titer for this antibody was 1:32. This sow's serum strongly agglutinated the erythrocytes of the foster pigs in vitro. Young and Underdahl (1949) estimated that the absorption of hemagglutination inhibition antibodies against swine influenza was complete in the baby pig within 24 hours. Their experiment was not designed to specifically determine this. Their estimate was based on samples drawn from the pigs at 24, 48, and 72 hours after birth. The samples showed no increase in titer after the 24-hour bleeding.

McGirr (1947) summarized the information available on the time after birth that various animal species were capable of absorbing intact antibodies. With the information summarized in Table 1, it would appear that domestic animals such as the calf, lamb, kid, foal, and pig, if allowed to nurse their dams for a few days after birth, would acquire the passive immunity in the form of antibodies which they would be capable of absorbing intact. However, a recent report by Petersen and Campbell (1955) has raised some doubt as to the validity of previous beliefs. In their report, two 5-month-old calves were each fed 6 liters of milk with a 1:1000 titer against Salmonella pullorum. No data were presented in their report, but the statement was made that the calves' blood promptly went from a negative to a positive agglutination test. In an experiment conducted on themselves and three graduate students who were negative to the test for Salmonella pullorum, positive titers up to a 1:10 dilution were detected within several days after consumption of one liter per day of 1:1000 titer milk. After two feedings of the milk, 2 adult pigs weighing 100 kilograms each were reported to react similarly with a positive test. Young and adult chickens shifted from negative to positive after feedings of immune milk. In guinea pigs and rabbits, the ingestion of 2 grams of the precipitated immune globulin resulted in the appearance of agglutinating antibodies in the blood.

There was some support for the work of Petersen and Campbell (1955), but these reports were substantiating for only some of the animal species. Cooledge (1916a) fed milk from Bacterium abortus infected cows to humans, and was able to detect agglutinins in their blood

sera. This was considered a passive immunity due to the absorption of the immune bodies in the milk. Walzer (1927) developed a method based on passive local skin sensitization to detect protein absorption in human subjects. The blood serum from a subject who was sensitized to a particular protein was introduced intradermally into another subject. When the test subject would eat this particular protein-containing food under specified conditions, a definite wheal developed at the sensitized site of intradermal injection. The two proteins tested were one raw egg and a 50-gram portion of raw herring, which were given to subjects in the morning before any other food was eaten. Lippard et al. (1936) criticized Walzer's (1927) work because the subjects had been fasted and sensitization could not be demonstrated in subjects on a normal diet. Lippard et al. (1936) performed 460 complement-fixation tests for the presence of lactalbumin in the blood sera of 229 normal human subjects ranging in age from 1 day to 30 years. Lactalbumin was found in infant's blood within a few days after ingestion of cow's milk. A positive test was present in 60 to 90 percent of the infants on the 4th to 20th day of age, but only isolated positive tests were observed after 5 months of age. Ratner and Gruehl (1934) used the skin sensitization method to determine whether medical students absorbed intact cottonseed proteins. They obtained the sensitized cottonseed serum from an asthmatic child sensitive to cottonseed. The majority of the students had a pronounced reaction at the site of intradermal serum injection within 15 to 30 minutes after consuming one teaspoon of dried cottonseed. Gruskay and Cooke (1955) worked with 21 infants whose ages ranged from 1 to 13

months and who were recovering from moderately severe diarrhea. The control group of infants and children, whose ages ranged from 5 days to 19 months, were convalescing from diseases unrelated to the gastrointestinal tract. None of the patients in either group had a history of previous egg ingestion. One gram of crystalline egg albumin per kilogram of body weight was administered orally after 4 to 8 hours of fasting. Venous blood samples were drawn 1 and 2 hours later. The amount of circulating egg albumin was determined by a quantitative precipitin reaction between the egg albumin antigen and rabbit anti-egg albumin serum. The concentrations of circulating egg albumin in the patients with diarrhea were significantly greater than in the controls. Egg albumin was demonstrated quantitatively in the sera of all control children and children suffering from diarrhea. The range of albumin concentration was 0.45 to 7.3 for the control children and 4.0 to 53.0 micrograms per milliliter of serum for the children suffering from diarrhea.

The mature guinea pig has been used quite extensively to show protein absorption because they readily develop signs of anaphylaxis after becoming sensitized to a foreign protein. Hartley (1942) used a collodian-particle agglutination method to demonstrate the absorption of crystalline egg albumin and the subsequent production of antibodies against this protein.

A report from the University of Wisconsin Agricultural Experiment Station (1957) refutes Petersen and Campbell's (1955) findings. These experiments with guinea pigs, rats, rabbits, dogs and monkeys all

suggest that only the newborn can effectively absorb antibodies in the blood stream from the intestinal tract. It was stated that adults probably cannot protect themselves from diseases by drinking milk containing antibodies which prevent these diseases.

## Possible Factors Causing the Reduction in Absorption of Antibodies from the Gastrointestinal Tract

In the early report of Smith (1925), faintly staining homogeneous bodies were found in the epithelial cells of the ileum of calves up to 3 days of age, and then these bodies disappeared. Comline et al. (1951a), made further detailed studies on this phenomenon. Cannulae were introduced into the duodenum, cecum and the thoracic duct of newborn calves which had not nursed. Then 500 milliliters of fat-free colostrum whey, with its globulins containing agglutinins against Brucella abortus, were introduced into the small intestine. Agglutinins were detected in the lymph, but not in the blood stream in 16 animals administered the colostrum whey fraction. These 16 calves were 6 to 27 hours old. Agglutinins were greatly reduced or absent in three calves 63 to 65 hours old. The same technique was used on four kids 8 to 21 hours old, and again agglutinins were found in the cannulated thoracic lymph duct secretion but not in the blood stream. The absorption of the unaltered immune globulins in the young animal entered the lymph system and was then carried to the peripheral blood system. In a later report Comline et al. (1951b) gives the results of the histological studies made on the same animals described above. They found the same type cells that Smith

(1925) had described in calves 9 to 36 hours old. Comparing histological samples from calves that had and had not been fed colostrum, they noted a striking difference in the intestinal epithelial cells. Globules with similar staining properties to colostrum globulins were found in the lumen of the gut and in the intestinal epithelial cells of the jejunum and ileum in those animals fed colostrum. In some instances, the globules could be seen in the lacteals and in the cytoplasm of the epithelial cells. Only a few droplets were seen in the histological sections in calves 63 to 65 hours old. Similar staining globules have been observed in colostrum-fed pigs and kittens, while no globules were found in the intestinal epithelial cells in animals 48 hours old that had not suckled (Comline et al., 1953). Hill and Hardy (1956) studied the histological changes in the lamb and kid, and also observed large numbers of eosinophilic globules in the epithelial cells of the villi and the lacteals in the newborn that had suckled. The globules were absent in the cells of older animals. Histochemical tests on the globules in the lumen, on the epithelial cells, and on the lacteals indicated that the globules were muco- or glycoprotein. It was reasonably certain that the globules were colostral proteins.

It has been assumed that immune globulins cease to be absorbed a few days after birth in domestic animals due to changes in the permeability of the intestinal mucosa. Hill (1956) presented experimental evidence whereby proteinaceous material might escape breakdown in the stomach. Three well-chosen animal species were studied. These were the rat, which was able to absorb antibodies for 20 days after birth, the

guinea pig which received most of its antibodies *in utero*, and the lamb, which was able to absorb antibodies for only 1 or 2 days. Histological studies and pH studies were made through the fetal stage and for a short time after birth. In the rat the peptic cells and parietal cells of the stomach were poorly developed up to 6 days after birth. At 12 and 18 days the glands were well defined, and histological samples contained moderate numbers of parietal cells and a few peptic cells. At 25 days the stomach mucosa resembled that of an adult rat. It was only after 20 to 25 days that a comparatively low pH was attained in the stomach contents of the rats. In the very young guinea pig, the pH of the stomach contents was between 1.0 and 2.0. The gastric glands were well defined in late fetal life, and by 1 hour after birth the gastric mucosa resembled that of the adult guinea pig. The 85-day fetal lamb had definite peptic cells with poorly-defined pepsinogen granules. The parietal cells were poorly developed, and continued to be widely scattered even at birth. The lamb at birth showed histological evidence for the secretion of two of the components of gastric juice, the pepsin and mucus, but lacked hydrochloric acid. A progressive increase in the number of parietal cells took place during the first 3 days of life, and at the end of 3 days the gastric glands resembled those of an adult. The pH of the stomach contents of the very young lamb was 5.0 to 7.0, the 24-hour-old lamb had a pH of 4.0, and the 36-hour-old lamb had a pH of 3.0. Hill (1956) cites the report of Hartley (1951) on the effect of peptic digestion on diptheria antitoxin and its failure to pass from the maternal blood stream to the fetal guinea pigs, although untreated diphtheria

antitoxin would. This was not because the antitoxin was destroyed, but because the enzyme activity had altered the structure. The cessation of antibody absorption in the newborn might be due to a comparable effect (Hill, 1956). Kastelic et al. (1950) postulated a pepsin and hydrochloric acid insufficiency in calves in his work on synthetic milk. Lewis (1956) reviewed the enzyme secretion of various animal species with respect to age. Hill (1956) also noted that an anti-trypsin factor had been found in bovine colostrum by Laskowski and Laskowski (1951) and Laskowski et al. (1952), and felt that this might aid in the absorption of unaltered immune globulins. It should be noted that Barrick et al. (1954) included the addition of trypsin inhibitor with orally administered porcine gamma globulin and bovine serum solids, but this addition did not effect the serum gamma globulin level in newborn pigs.

Halliday (1956) thought that the cessation of antibody absorption in the young rat might be initiated in rats and mice by eating solid food. Young rats prevented from taking solid food and compelled to live entirely on milk beyond the normal weaning age ceased to absorb antibodies at 20 days of age. Young rats fed on solid food at 12 days of age also continued to absorb antibodies until 20 days of age. It did not appear that solid food was responsible for bringing about the change in the permeability of the gut of the young rat to antibodies.

## Persistence of Passively Acquired Immune Globulins

Following the ingestion of colostrum by newborn domestic animals, a globulin of low mobility appears in the blood and increases rapidly in

concentration. This fraction with its immune bodies gives the young animal protection during the interim from birth until the animal's own antibody synthesizing mechanism becomes functional. Jameson et al. (1942) noted that the high concentration of gamma globulin present shortly after birth in calves decreased through the nursing period. McDiarmid (1946) followed the disappearance of colostral-derived Brucella abortus agglutinins in calves. The disappearance of agglutinins followed a logarithmic curve. The length of time which agglutinins persisted varied from a few days to a few months. The persistence depended to a large extent on the titer of the colostrum. Smith and Holm (1948) followed the disappearance of specific antibody activity as well as the change in the slow-moving, colostral-derived protein component in the calf. The half-lives for Hemophilus pertussis and vaccinia virus antibodies were approximately 50 days, and the half-life for diphtheria antitoxin was about 16 days. The estimation of the half-life of the blood serum slow-moving component, using the Tiselius apparatus, was 20 days.

McCarthy and McDougall (1953) observed that lambs with high titers of Salmonella typhosa antibodies acquired soon after birth fell to low values 5 or 6 weeks later.

Bangham and Terry (1957b) fed young rats adult rat serum labelled with radioactive iodine, $I^{131}$, and estimated the half-life of the selectively absorbed gamma globulin at 5.2 days.

Young and Underdahl (1949, 1950) studied the longevity of passively-conferred hemagglutination inhibitor antibodies in the baby pig. A

gradual decrease in titer took place from a few days after birth to 8 weeks of age. Foster et al. (1951) studied the composition of baby pig plasma from birth to 8 weeks of age. A decided decrease in the gamma globulin fraction occurred between the first day after birth and 3 weeks of age. There was a slight increase in gamma globulin between 3 and 8 weeks of age. No determinations were made between these ages.

Schoenheimer et al. (1942) and Heidelberger et al. (1942) studied the problem of persistence of antibodies both in passively and actively immunized rabbits. Using $N^{15}$ labelled amino acids as a tracer, the estimated half-life of an antibody molecule was about 2 weeks in actively immunized rabbits, but was only approximately 36 hours for the rabbits passively immunized with homologous pneumococcus Type I antiserum. In a rabbit, simultaneously actively and passively immunized, nitrogen uptake was found in the actively-produced antibody, while none was present in the passively acquired antibody.

Wiener (1951) used a factor to allow for the dilution resulting from increased body weight in babies to measure the half-life of Rh antibodies. The Rh antibody half-life determined from the logarithmic regression line was approximately 30 days. The author reported that a gamma globulin maximum was present at birth, and that this fraction diminished to its lowest point at 3 months of age. This was a slower decrease than would have been expected with a 30-day half-life.

Barr et al. (1949) studied the disappearance of diphtheria antitoxin in babies. The logarithmic regression line showed a rate loss of

13.9 percent, or approximately a 50 percent loss every 4.5 weeks in diphtheria antitoxin concentration.

## The Source of Antibodies in the Mammary Secretion

Following the report of Petersen and Campbell (1955), another paper by Campbell et al. (1957) described a phenomenon called "diathelic immunization," in which the milk from cows showed a positive agglutination titer against Salmonella pullorum a few hours after the cows had been nursed by calves exposed to this organism per os. The authors felt that the mammary gland functions as an exocrine reticuloendothelial gland, and that exposure to an antigen through the act of nursing causes an outpouring of a specific antibody in subsequent feedings. The importance of this phenomenon was tied with their previous contention that antibody absorption occurs via the intestinal tract for the duration of nursing and continues on into adulthood. The production of antibodies in the mammary gland would appear to be possible. Campbell et al. (1950) had shown earlier that plasma cells could be found in abundance in the mammary gland of the cow prior to parturition, shortly after parturition, and after experimental cessation of milking. Kolouch et al. (1947) from the same laboratory found an increase in plasma cells in the bone marrow of rabbits after antigen challenge.

Harris and Harris (1956) have reviewed the work relating the intimacy of the plasma cells with the production of antibodies. Cooledge (1916b) also reported the appearance of agglutinins in the milk against Br. abortus within 24 hours after the injection of pure culture of Br.

abortus into the udder of a cow. Blakemore (1947, 1951), to the contrary, was unable to stimulate local production of antibodies by the infusion of antigen (Br. abortus) into one-quarter of the udder of the cow. This treatment caused a rise in blood serum titer which was not reflected by the udder until 7 days after circulating antibodies appeared in the blood, and then the agglutinins appeared in all quarters simultaneously. Though Smith (1946), Smith et al. (1946), Smith and Coy (1946), and Smith and Greene (1947) have shown that the gamma globulin of the blood and the immune lactoglobulins differ quantitatively, the radiotracer work of Campbell et al. (1953), Askonas et al. (1954), and Blakemore and Garner (1956) leaves no doubt that antibody protein passes from the blood stream and becomes part of colostrum and milk immune proteins without degradation and resynthesis. This is not contradictory, since additions could take place to antibody molecules in their passage from the blood stream and through the mammary system. Supporting evidence for the blood protein gamma globulin as the source of immune lactoglobulin protein may be found in the reports of Friedell et al. (1951) in their studies on pregnant swine, and by Larson and Kendall (1957) in their studies on pregnant cows, which showed a definite decline in the gamma globulin of the serum protein prior to and immediately after parturition.

# EXPERIMENTAL

The experiments reported are on permanent file in the Swine Nutrition Section of the Animal Husbandry Department, Iowa State College, Ames, Iowa under the identical numbers used herein.

The following procedures and/or materials were common to two or more of the experiments reported herein. The pigs used in all of these experiments were from crossbred sows (Farmers Hybrid x Poland China x Landrace x Duroc) mated to Farmers Hybrid boars. The pigs were given an iron pill at three days of age, castrated at four days of age, and vaccinated with a mixture of hog cholera and erysipelas serum at five days of age.

The pigs were weighed at the initiation of the experiments and at weekly intervals thereafter, except in the experiments involving baby pigs during the first few days after birth.

Paper strip electrophoresis of the blood serum samples was carried out in a 3-compartment electrophoretic apparatus manufactured by the E-C Apparatus Company, 23 Haven Avenue, New York 32, New York, Serial No. 338. The procedure followed has been outlined by Raymond (1955) and has been included in Appendix A. The paper strips were developed by the staining technique described by Block et al. (1955). Details on this procedure have also been included in Appendix A. Optical density readings were obtained on each electrophoretic pattern on a densitometer manufactured by the Photovolt Corporation, New York. The relative concentration of each of the blood serum protein fractions were determined

by drawing a perpendicular beneath the trough between each fraction and measuring the area under the curve by planimetry.

All the statistical analyses made by analysis of variance procedures or t-tests were done as outlined by Snedecor (1956) and Cochran and Cox (1957).

The antigen-antibody immunological test employed in one of the phases of this study was adapted from the modified sheep red cell hemagglutination test described by Westphal et al. (1952a, 1952b) and modified by Neter et al. (1956). In this procedure the lipopolysaccharide fraction of Escherichia coli cells was extracted and absorbed on sheep red blood cells, and the modified erythrocytes were then used in the hemagglutination tests. This procedure has been outlined in Appendix B.

## Early Weaned Versus Sow Raised Pigs

### Objective

The objective of Experiment 764 was to determine if there was a difference in the relative concentration of the blood serum proteins in baby pigs weaned at one week of age compared to pigs that nursed their dams, both groups being carried on experiment until eight weeks of age. The growth rates of the two groups were also studied.

### Procedure

Two management procedures were followed in this experiment. The pigs from four crossbred litters of eight pigs each were grouped into littermate pairs based on their one-week weights. One pig from each of

the four pairs within each of the four litters was then randomly assigned to the early weaned group, while the other baby pig from each pair remained with the sow. At this time the sows and litters were moved from a heated central farrowing building, where they had been penned in farrowing stalls, to one of two four-pen wooden structure buildings. These buildings were not heated or insulated, but heat lamps were provided for the baby pigs of both groups as a supplemental heat source. The buildings had concrete floors, automatic water fountains, and self-feeders. In each of the two buildings, two of the sows and their four nursing pigs were penned separately, and in the other two pens the four pigs that were early weaned from each of these two sows were penned separately.

The sows were self-fed the lactation ration shown in Table 17 in Appendix C, from a few days before farrowing until the experiment was terminated when the pigs were 8 weeks old. The nursing pigs received no supplemental feed other than the sow ration which they began eating at approximately 5 weeks of age. Supplemental iron in the form of an iron pill was given to the nursing pigs at 1, 2, 3, 4 and 5 weeks of age. The weaned pigs were fed five pounds each of the prestarter diet shown in Table 13. This amount was enough for approximately one week. The diet was then changed to a starter ration (Table 15) and fed until the pigs were 5 weeks of age. A grower ration (Table 16) was then fed from 5 to 8 weeks of age.

Blood samples were drawn from the anterior vena cava of each of the 32 pigs at the initiation of this experiment, and subsequent samples

were obtained when the pigs were 2, 3, 4, 5, and 8 weeks of age. Approximately one ml. samples were withdrawn with 5 ml. syringes using 22 gauge needles for the first two bleedings and 19 gauge needles thereafter. The samples were then gently poured into 15 ml. blood centrifuge tubes and allowed to clot. After being placed in a refrigerator for approximately one hour, the samples were centrifuged at 2000 r.p.m. for 10 minutes, and the serum samples withdrawn by aspiration using 2 ml. pipettes. The serum samples were then placed in a refrigerator and held until the analyses were made, usually within 8 to 24 hours.

Paper strip electrophoretic separation of the serum proteins was then carried out. Two runs were made for each week's samples, and one determination for each sample was made in each run. Each separation was for a period of 10 hours, and at a potential of 150 volts and a current of 2.5 milliamps per paper strip.

Results and discussion

The results of this experiment are presented in Tables 2 to 6, inclusive, and in Figures 1 to 4, inclusive. The analysis of variance for each of the blood serum protein fractions are included in Table 20 in Appendix D. Only summary data dictated by a significance at P = 0.05 or less are included in this experiment.

The overall average relative percent of albumin, alpha globulin, beta globulin and gamma globulin varied significantly between the four litters, as did the average gains of the pigs. Significant differences in all measurements, except the beta globulin fraction was observed between the two treatments, and with the exception of the alpha globulin

Table 2. Experiment 764. Summary of the average relative percent of the blood serum protein fractions and pig gains by litter

| Litter | Albumin[a] | Alpha globulin[a] | Beta globulin[b] | Gamma globulin[a] | Gain/pig[b] |
|---|---|---|---|---|---|
| | | | | | (lb.) |
| 1 | 38 | 22 | 22 | 18 | 37.9 |
| 2 | 36 | 20 | 20 | 24 | 33.3 |
| 3 | 38 | 24 | 22 | 16 | 33.6 |
| 4 | 38 | 24 | 21 | 17 | 28.9 |

[a]Difference significant at P = 0.01 or less.

[b]Difference significant at P = 0.05 or less.

Table 3. Experiment 764. Summary of the average relative percent of the blood serum protein fractions and pig gains by treatment

| Treatment | Albumin[a] | Alpha globulin[a] | Beta globulin | Gamma globulin[b] | Gain/pig[a] |
|---|---|---|---|---|---|
| | | | | | (lb.) |
| On sows | 39 | 22 | 21 | 18 | 35.9 |
| Off sows | 36 | 23 | 21 | 20 | 30.9 |

[a]Difference significant at P = 0.01 or less.

[b]Difference significant at P = 0.05 or less.

Table 4. Experiment 764. Summary of the average relative percent of blood serum protein fractions and pig gains by weeks

| Weeks | Albumin[a] | Alpha globulin | Beta globulin[a] | Gamma globulin[a] | Gain/pig |
|---|---|---|---|---|---|
| | | | | | (lb.) |
| 1 | 34 | 23 | 22 | 21 | -- |
| 2 | 35 | 22 | 21 | 22 | 1.9 |
| 3 | 37 | 22 | 22 | 19 | 2.4 |
| 4 | 37 | 23 | 22 | 18 | 3.4 |
| 5 | 43 | 23 | 20 | 14 | 4.6 |
| 8 | 40 | 22 | 19 | 19 | 21.2 |

[a]Difference significant at P = 0.01 or less.

fraction, there was a significant difference between weeks. A comparison of the values obtained by averaging the litter values shown in Table 5, shows that the interaction of litter x weeks is significant for albumin, alpha globulin, gamma globulin, and average pig gains. Thus, the pig gains and certain serum protein fractions of the pigs in one litter behaved differently than pigs in another litter within the various weeks. The treatment x weeks interaction was also significant for all comparisons (Table 6). A clearer picture of this interaction can be obtained from Figures 1 and 2, where it was clearly evident that the two protein fractions, albumin and gamma globulin behaved differently within the various weeks tested. This also has been shown for the gamma

Table 5. Experiment 764. Summary of average relative percent of the blood serum protein fractions and pig gains by litter and weeks

| Litter | Week | Albumin[a] | Alpha globulin[a] | Beta globulin | Gamma globulin[b] | Gain/pig[b] (lb.) |
|---|---|---|---|---|---|---|
| 1 | 1 | 33 | 21 | 23 | 23 | -- |
| | 2 | 36 | 21 | 22 | 21 | 2.0 |
| | 3 | 39 | 21 | 22 | 18 | 2.6 |
| | 4 | 38 | 23 | 23 | 16 | 3.7 |
| | 5 | 43 | 23 | 21 | 13 | 5.1 |
| | 8 | 40 | 21 | 21 | 18 | 24.5 |
| 2 | 1 | 34 | 21 | 21 | 24 | -- |
| | 2 | 34 | 20 | 19 | 27 | 1.9 |
| | 3 | 36 | 20 | 19 | 25 | 2.6 |
| | 4 | 36 | 20 | 20 | 24 | 3.9 |
| | 5 | 39 | 20 | 20 | 21 | 4.6 |
| | 8 | 36 | 22 | 18 | 24 | 20.2 |
| 3 | 1 | 33 | 26 | 23 | 18 | -- |
| | 2 | 34 | 24 | 22 | 20 | 1.7 |
| | 3 | 36 | 25 | 24 | 15 | 2.0 |
| | 4 | 37 | 24 | 24 | 15 | 3.3 |
| | 5 | 45 | 24 | 20 | 11 | 4.9 |
| | 8 | 41 | 23 | 19 | 17 | 21.7 |
| 4 | 1 | 35 | 23 | 21 | 21 | -- |
| | 2 | 36 | 22 | 21 | 21 | 2.0 |
| | 3 | 38 | 23 | 22 | 17 | 2.1 |
| | 4 | 39 | 24 | 22 | 15 | 2.6 |
| | 5 | 43 | 24 | 21 | 12 | 3.8 |
| | 8 | 39 | 23 | 20 | 18 | 18.3 |

[a]Differences significant at P = 0.01 or less.

[b]Differences significant at P = 0.05 or less.

Table 6. Experiment 764. Summary of the average relative percent of the blood serum protein fractions and pig gains by treatment and weeks

| Treatment | Week | Albumin[a] | Alpha globulin[a] | Beta globulin[a] | Gamma globulin[a] | Gain/pig[a] |
|---|---|---|---|---|---|---|
| | | | | | | (lb.) |
| On Sows | 1 | 33 | 23 | 21 | 23 | -- |
| | 2 | 36 | 22 | 22 | 20 | 3.6[b] |
| | 3 | 39 | 21 | 23 | 17 | 3.2[b] |
| | 4 | 41 | 21 | 23 | 15 | 4.2[b] |
| | 5 | 46 | 21 | 20 | 13 | 4.4 |
| | 8 | 39 | 22 | 19 | 20 | 20.5 |
| Off sows | 1 | 34 | 23 | 23 | 20 | -- |
| | 2 | 34 | 22 | 20 | 24 | 0.2[b] |
| | 3 | 35 | 23 | 21 | 21 | 1.5[b] |
| | 4 | 34 | 25 | 21 | 20 | 2.6[b] |
| | 5 | 39 | 25 | 20 | 16 | 4.8 |
| | 8 | 39 | 22 | 20 | 19 | 21.8 |

[a]Differences significant at P = 0.01 or less.

[b]Differences significant at P = 0.05 or less.

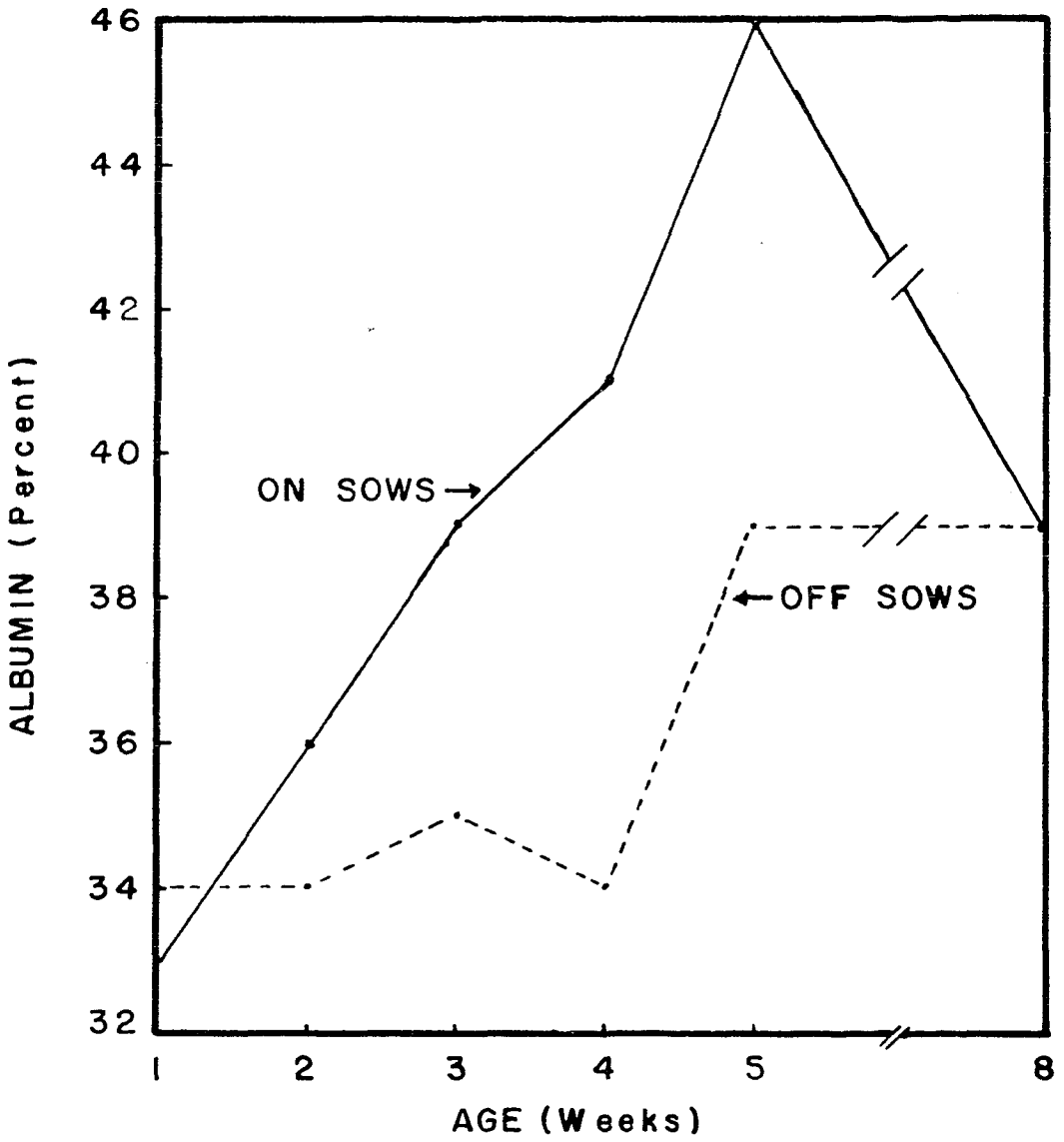


Figure 1. Experiment 764. The relative percent of the albumin blood serum protein fraction versus age.

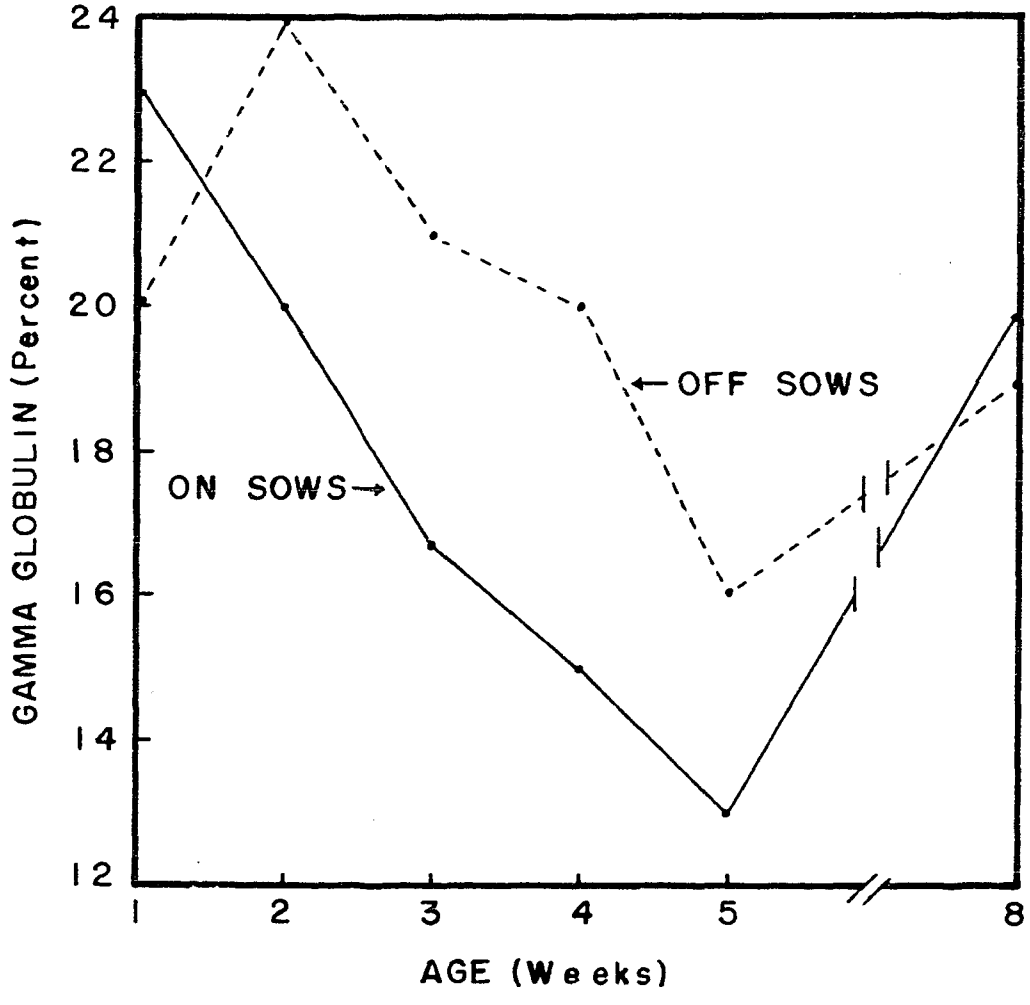


Figure 2. Experiment 764. The relative percent of gamma globulin blood serum protein fraction versus age.

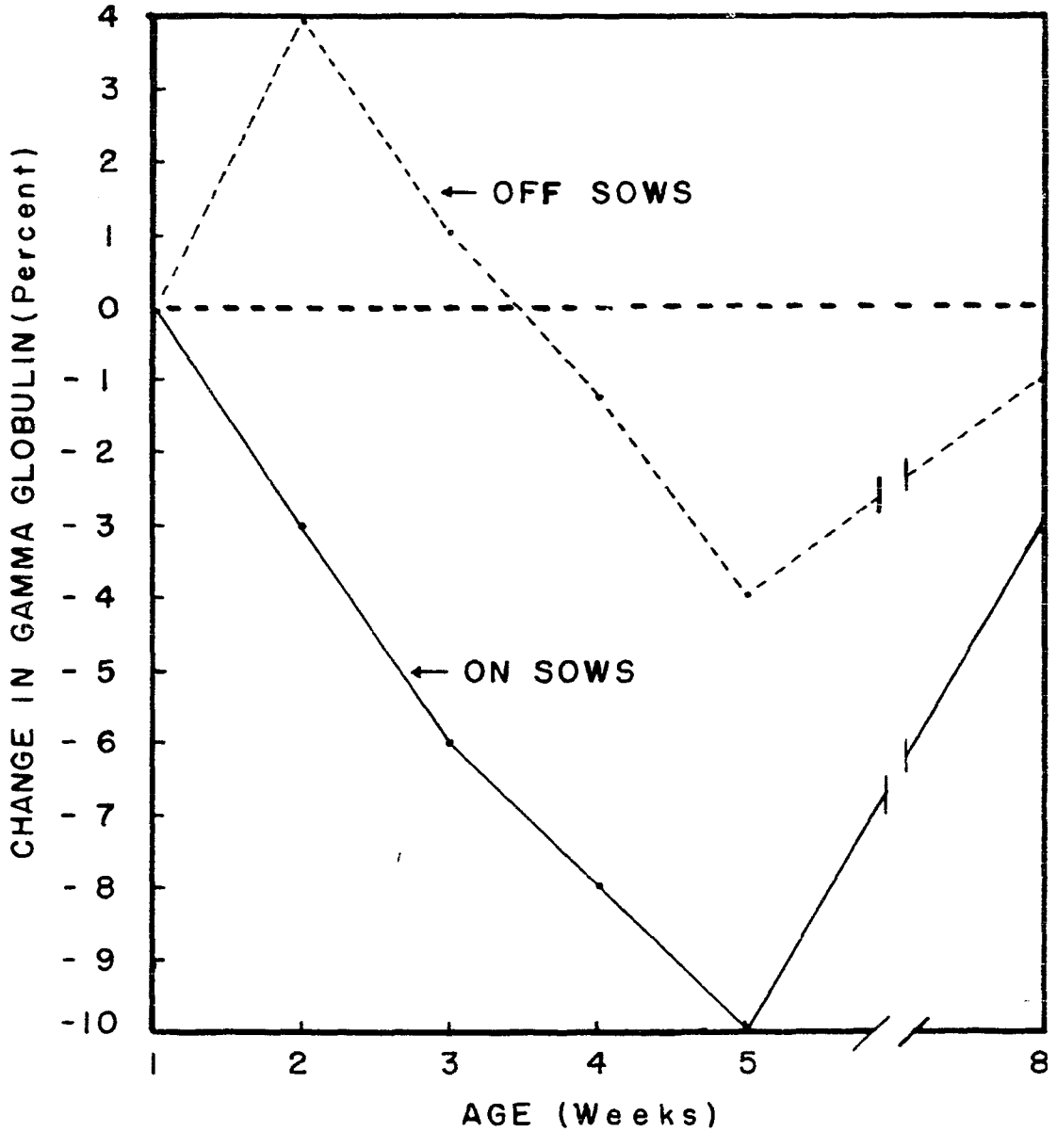


Figure 3. Experiment 764. The percentage unit change in the gamma globulin blood serum protein fraction versus age.

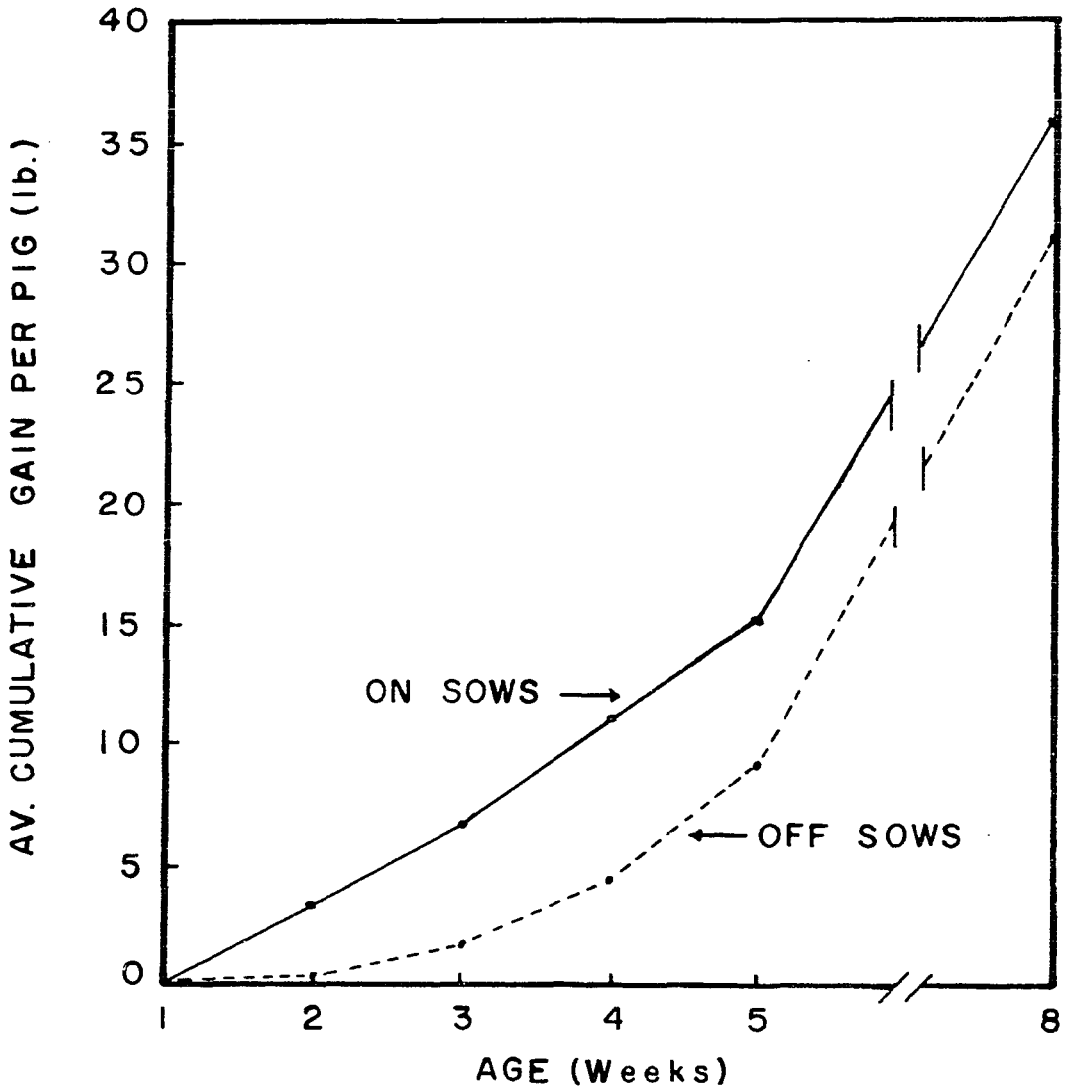


Figure 4. Experiment 764. Growth curves of baby pigs versus age.

globulin in Figure 3, where the percentage unit change from week to week has been plotted.

During the periods 1 to 2, 2 to 3 and 3 to 4 weeks of age, the pigs being nursed by their dam gained significantly more than the pigs early weaned. However, between the 4 to 5, and 5 to 8 week period the early weaned pigs gained slightly more than the sow raised pigs, but the advantage was not significantly different. The early weaned pigs finished the experiment 5 pounds lighter on the average than their littermate pairs who were nursing their dams.

Of particular interest in this experiment was the changes in the gamma globulin level of the blood serum in the two groups of pigs. Before the experiment was carried out, it was suspected that there probably would not be any differences, or if any, it was thought that possibly the pigs nursing their dams might have a slightly higher level of serum gamma globulin. On the contrary, the apparent rise in gamma globulin level in the early weaned pigs was surprising. This phenomenon was undoubtedly due to the fact that the early weaned pigs gained so poorly the first week after weaning, and as a result of what appears to be a higher catabolism of the alpha and beta globulin fractions, the relative gamma globulin content showed an increase. The fact that the increased gamma globulin level in pigs from two of the litters was enough to cause an increase in the litter gamma globulin level at the end of the first week accounted for the significant interaction in the litters x weeks interaction. After this second week the trend for the rest of the weeks in all the litters was very similar.

The better growth of the nursing pigs can be attributed in part to the milk production of the sows with respect to the relatively few number of pigs that were nursing. In addition one sow had five functional teats, one sow six, and two sows had seven teats that were producing milk at the end of the experiment. In each case there was more than one functional teat per pig for the nursing pig group. Also, the non-insulated wooden buildings that housed the early weaned pigs and the sows and litters would be classified as sub-standard for early weaning baby pigs, especially for winter operation.

## Antibody Absorption by the Newborn Pig

### Objective

The objective of Experiment 778 was to determine the length of time after birth that the newborn baby pig was able to absorb an antibody from the intestinal tract and into the blood stream, when the antibody was given *per os*.

### Procedure

The modified sheep red cell hemagglutination test previously mentioned and outlined in detail in Appendix B, was used as the criterion to measure antibody absorption. The test was also used to quantitatively measure the efficiency of antibody absorption in baby pigs of various ages.

Sow colostrum which contained an antibody against *E. coli* was used as the dosage material to measure the absorption and efficiency of

absorption of antibodies by baby pigs. Standardized E. coli antigen (killed organisms) was injected, both subcutaneously and intravenously, into three groups of sows several weeks before farrowing. The injection rates and dosage schedule for these 38 sows have been summarized in Table 25 in Appendix E. When these sows farrowed, their baby pigs were removed from the farrowing stall within 12 hours after the sow farrowed. In order to allow some time for the colostrum to accumulate, the sow was not milked until approximately three hours after the pigs had been removed. One or two ml. of oxytocin solution containing 10 I.U. per ml. (Pitocin, Parke Davis and Company, Detroit, Michigan) was injected intravenously via an ear vein into each sow and the colostrum was expressed manually from the teats of the sow. The hemagglutination titers of the colostrum samples so obtained have been shown in Figures 7, 8, and 9 in Appendix E.

The baby pigs used in this experiment were farrowed by non-challenged (low-titer) sows, and were allowed to nurse their dams naturally except when they were removed only long enough to obtain a blood sample and to dose them with high-titer colostrum. The sows were all fed the lactation ration shown in Table 17 in Appendix C. Blood samples were obtained from the anterior vena cava of the baby pigs, and the blood was handled in the same manner as previously described. One or more pigs were used as control pigs in each litter to obtain an estimate of the antibody absorption obtained from their dam who invariably showed a low titer in their colostrum against the E. coli antigen used in this experiment. In addition to these control pigs, each pig dosed with high-titer

colostrum served as its own control, since a blood sample was always drawn just prior to dosing the pig. Reciprocal titers were recorded in each case, and represented the highest dilution at which hemagglutination was evident. All dilutions of 1:10 which did not show positive hemagglutination were recorded as zero (0).

Three litters of pigs were selected in Group I for this phase of the experiment. Four pigs were selected at the time they farrowed or very shortly after farrowing. Blood samples were drawn and two of these pigs then orally dosed with high-titer colostrum using a stomach tube and calibrated syringe. The oral dosage rate was 20 ml. of colostrum per kg. of body weight. One of the pigs orally dosed was dosed again 12 hours later, and then blood samples were obtained from the four pigs 12 hours later (24 hours of age). Two additional pigs from each litter were randomly selected at 12, 24, 48, and 72 hours after farrowing. Immediately after a blood sample was drawn, the pigs were orally dosed with high-titer colostrum and 12 hours later one pig from each of the above pairs was again orally dosed. Both pigs from each of the time intervals were bled a second time (24 hours after dosing began).

A blood sample was drawn from each of the 34 pigs 168 hours (7 days) after farrowing. At this time the two control pigs from each litter were orally dosed, one of these received another dose 12 hours later, and another blood sample drawn from each 12 hours later.

Two composited batches of colostrum were used for oral dosage on this group of pigs. One batch used to orally dose the pigs from 0 to the 24 hour time periods, inclusive, showed a positive hemagglutination at

the 1:320 dilution. The other batch agglutinated at a dilution of 1:640 and was used to orally dose the pigs from the 48 hour time period to the end of the test.

The procedure followed on the Group II pigs was similar to the previous group, with the exceptions noted below.

Five litters of pigs were employed in this phase of the experiment. Since the previous group of pigs had shown little antibody absorption beyond the 0-hour group, the dosage rate of the colostrum was doubled for this group. One of the treatment pigs from each litter and at each time interval received 20 ml. of colostrum per kg. of body weight in each of four dosages. These dosages were at 6-hour intervals starting immediately after the initial blood sample was drawn. The other pig from each litter and each time interval received the same amount of colostrum per unit of weight in each of two dosages. These dosages were at 12-hour intervals starting immediately after the initial blood sample was drawn. Oral dosage was started on two pigs in each litter at 0, 24 and 48 hours after birth. An initial blood sample was obtained from each pig, and then two additional samples at 12-hour intervals. Oral dosage of the colostrum always followed immediately after the blood sample was drawn.

The titer of the composited colostrum used in this experiment varied, but is included for each pig in Table 8.

In Group III one baby pig was selected from each of eight litters for oral dosage of high-titer colostrum every 6-hour interval beginning at 0 hour through 36 hours after farrowing. Blood samples were also

obtained from two control pigs in each litter, one selected at 0 hour, and one selected at 48 hours after birth. Unless it was a control pig, each pig was given one oral dose of high-titer colostrum at the rate of 40 ml. per kg. of body weight immediately following the initial bleeding. All pigs were again bled 12 hours after the initial blood sample was obtained. Immediately after this blood sample was drawn, the pigs were given an injection of colostrum whey fraction intraperitoneally at the rate of 10 ml. per kg. of body weight, and the pigs bled again 12 hours later.

Colostrum composites of two different agglutinating potencies were used in this phase of the experiment. Colostrum that agglutinated modified red cells at a dilution of 1:10,240 was used to dose the pigs in litter numbers 9 to 14, inclusive, and the titer of the colostrum used to dose the pigs in litter numbers 15 and 16 was positive at the 1:20,480 dilution. The colostrum whey had the same hemagglutination titer that the whole colostrum did for each litter as outlined above.

Results

The initial reciprocal hemagglutination titer and the titer obtained 24 hours later, for each of the serum samples drawn from the baby pigs in Group I, have been shown in Table 7. This summary includes all the data, except for the titers obtained at 168 hours for the pigs that were dosed at 0, 12, 24, 48, and 72 hours. These titers were in each case the same or lower than the titer obtained on the second bleeding samples. Only those pigs orally dosed at birth (0 hour) gave any indi-

cation of absorbing this specific E. coli antibody. There appeared to be little or no difference in dosage rate.

The results obtained from the Group II pigs have been summarized in Table 8. Antibody absorption was definitely shown to have taken place when colostrum dosage was started immediately after birth (0 hour). The higher dosage rate resulted in higher serum titers in three out of the five comparisons made 12 hours after farrowing, but in only two cases in the comparison of the same pigs 24 hours after farrowing. Oral dosage of high-titer colostrum, when started after the pigs had reached an age of 24 hours, yielded questionable results.

The results of the serum hemagglutination tests run on the Group III pigs have been presented in Table 9. Antibody absorption resulting from the oral dosage of high-titer colostrum was clearly evident for the several hours after birth. Since the data were available for each pig, a paired comparison t-test was made between the initial and subsequent 12-hour blood serum sample obtained after the oral dosage of colostrum. This test was made for each of the time interval groups. The difference between the initial and subsequent blood serum titers of the pigs for the 0-, 6-, 12-, and 18-hour groups was significant at $P = 0.05$ or less. Differences observed in the pigs of the 24-, 30-, and 36-hour groups were not significant. The average differences for the various 6-hour interval groups have been shown in Figure 5.

The results obtained by the injection of the colostrum whey fraction have been shown in Table 9.

Table 7. Experiment 778 (Group I). Serum hemagglutination titers of baby pigs

| Control sow number and colostrum titer | | Control pigs Hrs. after birth | | Treatment | Treated pigs[a] Hours after birth | | | | | | | | | | | |
|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|
| | | 0[b,c,d] | 24 | | 0[b,c,d] | 24 | 12[b,c] | 36 | 24[b,c] | 48 | 48[b,c] | 72 | 72[b,c] | 96 | 168[b,c] | 192 |
| 1. | 80 | 10 | 20 | 1 | 10 | 40 | 20 | 20 | 40 | 20 | 0 | 0 | 0 | 10 | 0 | 0 |
| 1. | 80 | 0 | 40 | 2 | 0 | 80 | 40 | 20 | 20 | 10 | 10 | 10 | --[e] | --[e] | 20 | 0 |
| 2. | 160 | 10 | 80 | 1 | 0 | 80 | 80 | 80 | 10 | 20 | 40 | 80 | 40 | 40 | 0 | 10 |
| 2. | 160 | 0 | 80 | 2 | 0 | 80 | 80 | 80 | 40 | 40 | 40 | 40 | 10 | 20 | 10 | 20 |
| 3. | 40 | 10 | 10 | 1 | 0 | 80 | 20 | 0 | 20 | 20 | 0 | 0 | 0 | 0 | 0 | 0 |
| 3. | 40 | 0 | 20 | 2 | 10 | 40 | 10 | 0 | 10 | 0 | 10 | --[f] | --[e] | --[e] | 0 | 0 |

[a]Colostrum with 1:320 titer was used to orally dose all treatment pigs from 0 to 24 hours time periods inclusive, and colostrum with 1:640 titer used from 48 hours to end of the test.

[b]Treatment 1 was 20 ml. colostrum per kg. of body weight, and treatment 2 was two times 20 ml. colostrum per kg. of body weight.

[c]Initial blood sample drawn and then the pig orally dosed, unless it was a control pig.

[d]Some pigs nursed for short period before serum sample drawn.

[e]Not enough pigs in the litter to obtain these data.

[f]Pig was overlain by the sow.

Table 8. Experiment 778 (Group II). Serum hemagglutination titers of baby pigs

| Control sow no. & colostrum titer | Control pigs: Hours after birth 0[a] | Control pigs: Hours after birth 24 | Treatment[b] | Pigs orally dosed with colostrum: Hours after birth 0[a] | 12 | 24 | Treatment colostrum titer | Hours after birth 24[a] | 36 | 48 | Treatment colostrum titer | Hours after birth 48[a] | 60 | 72 | Treatment colostrum titer |
|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|
| 4. 40 | 0 | 20 | 1 | 0 | 160 | 40 | 640 | 10 | 40 | 40 | 160 | 80 | 80 | 40 | 160 |
| 4. 40 | 0 | 20 | 2 | 0 | 80 | 40 | 640 | 10 | 40 | 40 | 160 | -[c] | -[c] | -[c] | - |
| 5. 40 | 0 | 0 | 1 | 0 | 160 | 80 | 1280 | 10 | 20 | 20 | 1280 | 20 | 40 | 40 | 10,240 |
| 5. 40 | 0 | 0 | 2 | 0 | 80 | 80 | 1280 | 0 | 40 | 20 | 1280 | 10 | 40 | 40 | 10,240 |
| 6. 160 | 0 | 20 | 1 | 0 | 320 | 320 | 1280 | 80 | 40 | 40 | 1280 | 40 | 40 | 40 | 10,240 |
| 6. 160 | 0 | 40 | 2 | 0 | 320 | 320 | 1280 | 40 | 20 | 40 | 1280 | 40 | 40 | 40 | 10,240 |
| 7. 80[d] | 10 | 40 | 1 | 0 | 640 | 1280 | 10,240 | 10 | 0 | 0 | 10,240 | 0 | 0 | 0 | 10,240 |
| 7. 80[d] | 10 | 40 | 2 | 10 | 640 | 640 | 10,240 | 0 | 10 | 10 | 10,240 | 20 | 10 | 0 | 10,240 |
| 8. -[e] | 0 | 0 | 1 | 0 | 1280 | 640 | 10,240 | 0 | 0 | 0 | 10,240 | 0 | 0 | 0 | 10,240 |
| 8. -[e] | 0 | 0 | 2 | 0 | 320 | 320 | 10,240 | 0 | 0 | 0 | 10,240 | 0 | 0 | 0 | 10,240 |

[a]Initial blood sample drawn at this hour and then the pig orally dosed, unless it was a control pig.

[b]Treatment 1 was four doses of 20 ml. colostrum per kg. of body weight each and treatment 2 was two doses of 20 ml. colostrum per kg. of body weight.

[c]Not enough pigs in the litter to obtain these data.

[d]The pigs farrowed in this litter had nursed for a short time before the initial blood samples were drawn.

[e]The pigs in this litter were obtained by Caesarean section. The sow died and the pigs were transferred to another sow.

Table 9. Experiment 778 (Group III). Serum hemagglutination titers of baby pigs

| Control sow number and colostrum titer | | Control pigs | | | Treated pigs | | | | | |
|---|---|---|---|---|---|---|---|---|---|---|
| | | Hours after birth | | | | | | | | |
| | | 0[a] | 12[b] | 24 | 0[a] | 12[b] | 24 | 6[a] | 18[b] | 30 |
| 9. | 10 | 0 | 0 | 320 | 0 | 640 | 640 | 0 | 320 | 320 |
| 10. | 0 | 0 | 10 | 1280 | 0 | 2560 | 5120 | 10 | 640 | 2560 |
| 11. | 10 | 0 | 0 | 640 | 0 | 2560 | 2560 | 10 | 320 | 1280 |
| 12. | 40 | 0 | 0 | 5120 | 0 | 20,480 | 10,240 | 0 | 5120 | 640 |
| 13. | 20 | 0 | 20 | 1280 | 0 | 5120 | 10,240 | -[c] | 2560 | 5120 |
| 14. | 160 | 0 | 20 | 640 | 0 | 5120 | 1280 | 0 | 1280 | 1280 |
| 15. | 40 | 0 | 0 | 640 | 0 | 2560 | 2560 | 0 | 320 | 1280 |
| 16. | 80 | 0 | 40 | 640 | 0 | 2560 | 2560 | 0 | 2560 | 2560 |

| | | Treated pigs | | | | | | | | |
|---|---|---|---|---|---|---|---|---|---|---|
| | | Hours after birth | | | | | | | | |
| | | 12[a] | 24[b] | 36 | 18[a] | 30[b] | 42 | 24[a] | 36[b] | 48 |
| 9. | 10 | 0 | 640 | 640 | 0 | 0 | 640 | 0 | 20 | 320 |
| 10. | 0 | 10 | 80 | 320 | 0 | 40 | 640 | 10 | 40 | 320 |
| 11. | 10 | 0 | 0 | 640 | 0 | 40 | 5120 | 10 | 0 | 2560 |
| 12. | 40 | 10 | 40 | 2560 | 0 | 10 | 5120 | -[d] | 20 | 1280 |
| 13. | 20 | 40 | 640 | 320 | 20 | 80 | 320 | 0 | 40 | 640 |
| 14. | 160 | 0 | 320 | 320 | 20 | 80 | 640 | 10 | 10 | 320 |
| 15. | 40 | 0 | 160 | 1280 | 0 | 40 | 1280 | 0 | 0 | 1280 |
| 16. | 80 | 20 | 160 | 2560 | 10 | 40 | 640 | 10 | 40 | 1280 |

[a]Time of initial bleeding. Treated pigs then immediately orally dosed with high-titer colostrum.

[b]Time of 2nd bleeding and high-titer colostrum whey then injected intraperitoneally.

[c]This sample was lost.

[d]The serum hemolyzed the modified red cells.

Table 9 (Continued)

| Control sow number and colostrum titer | | Treated pigs | | | | | | Control pigs | |
|---|---|---|---|---|---|---|---|---|---|
| | | Hours after birth | | | | | | | |
| | | 30[a] | 42[b] | 54 | 36[a] | 48[b] | 60 | 48[e] | 60 |
| 9. | 10 | 0 | 40 | 640 | 10 | 20 | 640 | 0 | 160 |
| 10. | 0 | 10 | 40 | 1280 | 0 | 0 | 2560 | 10 | 640 |
| 11. | 10 | 20 | 20 | 2560 | 0 | 0 | 2560 | 20 | 2560 |
| 12. | 40 | 0 | 0 | 1280 | -[d] | -[d] | 640 | 0 | 1280 |
| 13. | 20 | 20 | 0 | 320 | 10 | 0 | 640 | 0 | 640 |
| 14. | 160 | 10 | 10 | 1280 | 10 | 0 | 640 | 10 | 1280 |
| 15. | 40 | 0 | 0 | 320 | 0 | 10 | 1280 | 10 | 640 |
| 16. | 80 | 20 | 20 | 2560 | 40 | 20 | 640 | 20 | 1280 |

[e]Time of first bleeding on 48-hour-old control pigs and high-titer colostrum whey then injected intraperitoneally.

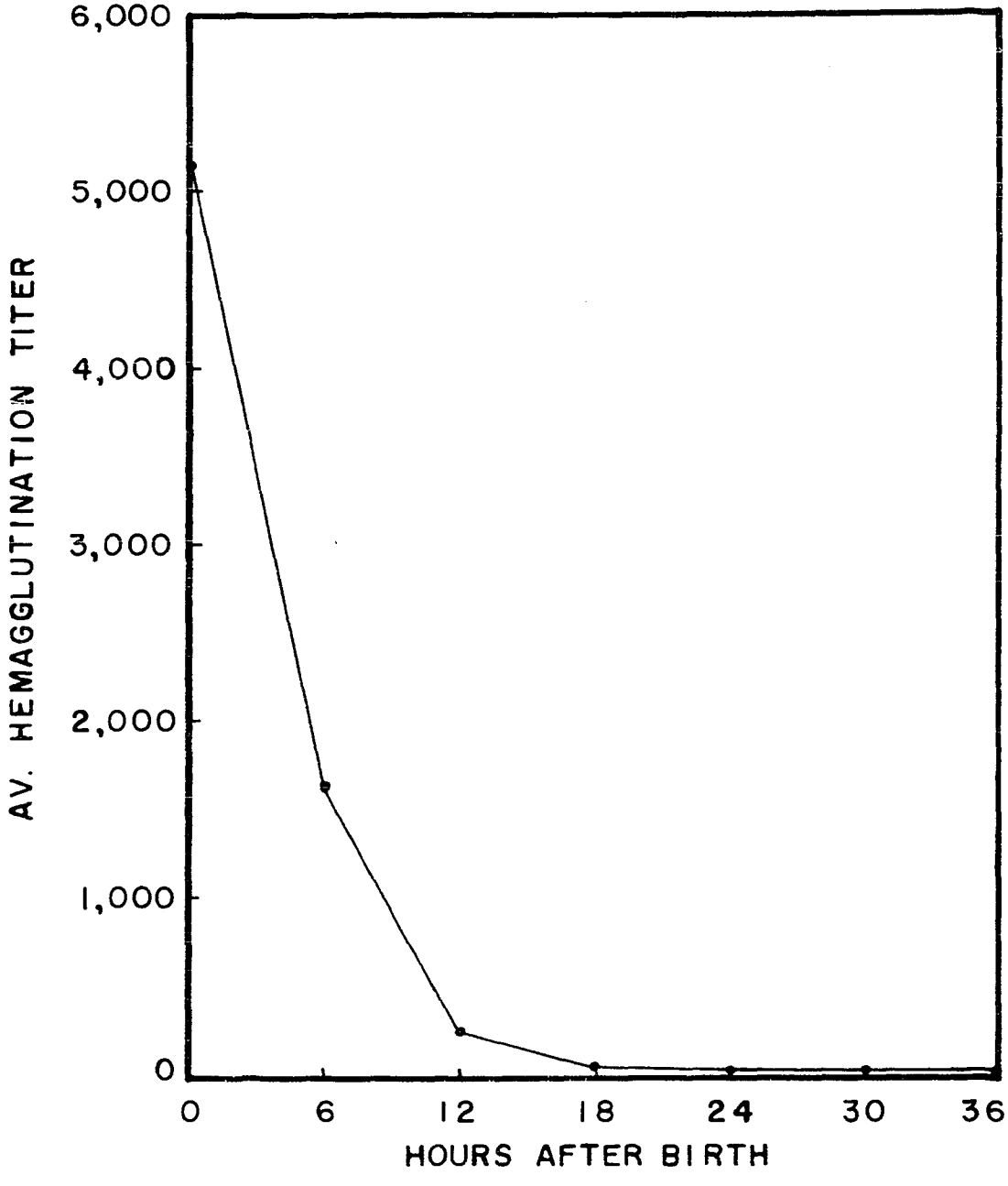


Figure 5. Experiment 778 (Group III). The average difference between the initial blood serum hemagglutination titer and the titer obtained 12 hours after the oral dosage of high-titer colostrum versus the hours after birth or the time-interval the pigs were dosed.

Discussion

In this experiment, the pigs in Group I and in Group II provided evidence that the baby pig's ability to absorb a specific antibody was short-lived, and probably lasts no longer than 24 hours after birth. Since the colostrum used in some instances had a titer that was not as high as might have been desirable, further data were needed to more accurately determine answers to the objectives being pursued. It was also possible that the specific E. coli antibody might have been absorbed, and was escaping detection by the hemagglutination test. The antibody may have been absorbed in the 24-hour-old pigs or older, and possibly metabolized or adsorbed in the lymph masses or reticuloendothelial system.

From the data obtained from the pigs in Group III, the ability to absorb an antibody by the young pig was readily discernible by the hemagglutination test. Further, this ability appeared to decrease rapidly within a few hours. In an attempt to statistically analyze these data to segregate the regression components of the variation due to the differences in the time interval groupings, it was evident that the data lacked homogeneity of variances. This limited the interpretation and reliability of the analysis of variance. Proceeding further, the reciprocal hemagglutination titers observed 12 hours after oral colostrum dosage in each time group interval were transformed to logarithmic values. Since there were a considerable number of zero (0) values, it was decided to arbitrarily assign a value of ten (10) to these. Tests of homogeneity on these data revealed they were still heterogeneous. Further tests were made by dropping out the 36-hour interval group, and then the 30-hour

group. When the 30-hour interval group was eliminated, the remaining logarithmic data proved to be homogeneous. A significant linear regression (P = 0.01 or less) was found to fit these data when an analysis of covariance was made. This analysis will be found in Table 21 in Appendix D. The regression line was fitted to the data and has been shown in Figure 6. The antibody absorption half-life was determined using the first order reaction rate equation where $t_{\frac{1}{2}} = \frac{0.693}{\lambda}$ and where lambda ($\lambda$) = -0.227. The half-life was found to be equal to 3.06 hours, or a 50 percent reduction occurred in antibody absorption in this time.

The intraperitoneal injection showed that if the antibody had been absorbed via the intestinal tract by the 24-, 30-, or 36-hour-old pigs with any moderate degree of efficiency, it would have been detected by the hemagglutination test. Note that in the 30- and 36-hour interval groups of pigs, the average titer found after the injection of the high-titer whey was very close to one-fourth the average value of the 0-hour group after oral dosage of colostrum. Since the dosage rate of the whey injection was one-fourth that orally dosed, this would mean nearly 100 percent absorption of the antibody had taken place at birth by the pigs.

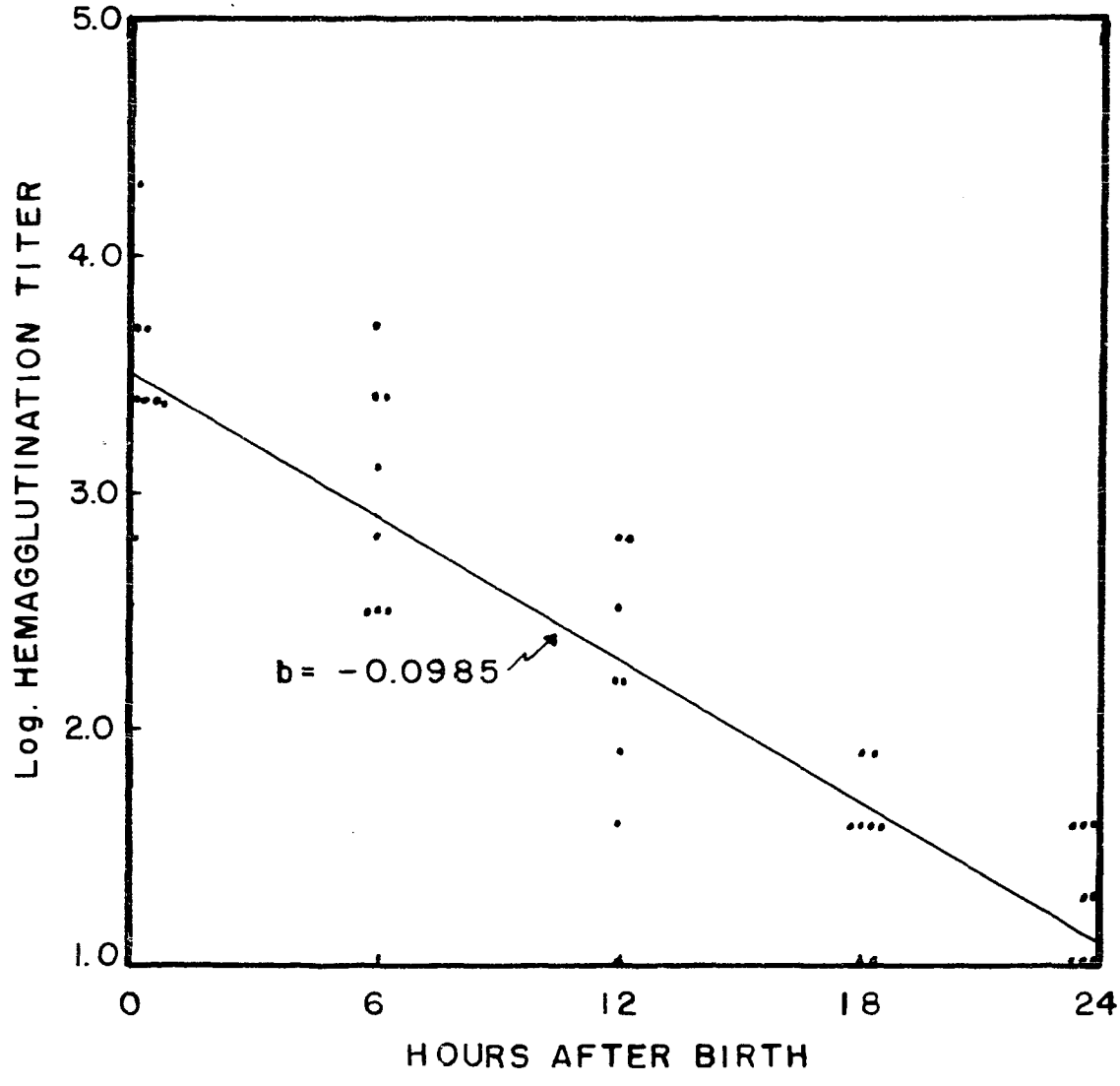


Figure 6. Experiment 778 (Group III). Logarithmic values of the hemagglutination titers versus the hours after birth or the time-interval the pigs were dosed.

# Effect of Supplemental Colostrum, Porcine Gamma Globulin, and Other Factors on the Blood Serum Proteins and Growth of Early Weaned Pigs

## Objectives

The objectives of these three experiments were to determine whether supplemental colostrum whey fraction, porcine gamma globulin, hog cholera antiserum or a high molecular weight polysaccharide would have any effect on the blood serum protein fractions, particularly the gamma globulin fraction. It was also desirable to know whether these treatments would have any effect on the growth of early weaned pigs and whether there was a correlation between the gamma globulin protein fraction and the growth rate.

## Procedures

Several of the management procedures in these three experiments were the same. In all of the experiments the baby pigs were weaned at approximately 7 days of age. The pigs had previously been given an iron pill, injected with 10 ml. of a mixture of hog cholera and swine erysipelas antiserum, and all males were castrated. All the pigs were fed an average of five pounds of the prestarter diet shown in Table 13 in Appendix C. This amount was enough for each pig for approximately the first week after being weaned. The pigs were weighed at the initiation of the experiments and at weekly intervals thereafter. The pens were cleaned daily and shavings were provided for bedding, except for the first two days when the experiments were started. No bedding was supplied at this

time because it was thought the pigs learned to eat the dry prestarter sooner. The self-watering fountains were flushed daily, and the self-feeders were inspected twice daily to be certain that ample feed was always available.

The gamma globulin used in these experiments was a 10 percent Porcine Gamma Globulin Solution, Lot Number R 527051 from the Armour Veterinary Laboratories, Armour and Company, Chicago, Illinois. The colostrum was obtained from sows in the Iowa State College Swine Nutrition herd. It was collected during previous farrowing as previously described, and was immediately frozen and stored until ready for use. The whey fraction was separated as described in Appendix B. The high molecular weight polysaccharide, Zymosan, was obtained from Standard Brands, Incorporated, New York 22, New York. The hog cholera antiserum was obtained from Allied Laboratories, Incorporated, Indianapolis, Indiana.

Experiment 723

Nine treatments were compared in a 3 x 3 balanced lattice experimental design in this experiment. Four pigs were individually fed per treatment. The initial weight and age of the pigs were 6.4 pounds and 9.4 days, respectively. Littermate groups of three pigs each made up the 12 outcome groups. After the five pounds of prestarter fed each pig was consumed, they were then fed the starter ration shown in Table 14 in Appendix C.

The building used to house the pigs for this experiment contained 36 individual pens. The pens were arranged on a concrete slab which was radiantly heated by circulating hot water from a thermostatically con-

trolled hot water heater. The building temperature was controlled with an air conditioning, forced-air gas furnace combination unit. Germicidal lamps surrounding the walls of this building were on continuously. The air was positively exhausted continuously and all air entering the building passed through a bank of germicidal lamps. The microbial contamination in this building was undoubtedly lower than other buildings located at the Swine Nutrition Farm.

Additions that were made to the basal or control regime has been shown in Table 10. These additions were made when the pigs were started on the experiment and at weekly intervals thereafter for the first, second and third week, making a total of four supplementations. The injections were given intraperitoneally. In those treatments where the additions were fed, the material was mixed with a small amount of the ration and placed in the feeder. This was generally consumed within 12 to 18 hours.

Blood samples were drawn and electrophoretic analyses of the serum was made on each pig, as previously described, when the experiment terminated after 4 weeks. The electrophoretic separations were carried out for a period of 4 hours at a potential 460 volts and a current of 10 milliamps per strip.

Experiment 734

Five treatments were compared in this experiment. A randomized block design was used in the allotment of littermate outcome groups of 5 pigs each from a total of 20 litters. Because of limited pen space, the littermate outcome groups of pigs in this experiment were penned together.

Table 10. Experiment 723. Summary of the gain, feed per pound of gain, and the relative percent of the blood serum protein fractions

| Treatment (weekly) | Gain/pig (lb.) 1-5 wks. | Feed/lb. gain | Relative percentage of blood serum proteins | | | |
|---|---|---|---|---|---|---|
| | | | Albumin | Alpha globulin | Beta globulin | Gamma globulin |
| Basal | 15.6 | 1.78 | 37 | 29 | 22 | 12 |
| Basal + 10 mg. Zymosan injected | 11.9 | 1.96 | 38 | 27 | 24 | 11 |
| Basal + 100 mg. Zymosan injected | 11.2 | 1.84 | 39 | 27 | 22 | 12 |
| Basal + 100 mg. Zymosan fed | 10.9 | 2.02 | 36 | 28 | 24 | 12 |
| Basal + 30 ml. porcine gamma globulin injected | 10.8 | 2.06 | 33 | 28 | 24 | 15 |
| Basal + 60 ml. porcine gamma globulin fed | 14.9 | 1.88 | 38 | 27 | 24 | 11 |
| Basal + 15 ml. sow colostrum whey fraction injected | 10.7 | 2.02 | 34 | 28 | 26 | 12 |
| Basal + 30 ml. sow colostrum whey fraction fed | 12.4 | 1.86 | 36 | 28 | 24 | 12 |
| Basal + 40 ml. hog cholera serum injected | 14.3 | 1.70 | 36 | 29 | 22 | 13 |

Thus, feed records, though they were obtained, were on a litter basis and could not be separated for treatment comparisons. After the pigs had consumed an average of five pounds each of the prestarter ration, they were fed the starter ration shown in Table 14 in Appendix C. In this experiment, the pigs were not allotted to the treatments until one week after they had been weaned. At this time the average weight and age was 7.6 pounds and 15.7 days, respectively. The experimental period was for three weeks. The treatment additions were given at the beginning and after the first and second weeks the pigs were on experiment. These treatments have been shown in Table 11. The amount of each material administered to each pig was 5 ml. per kg. of body weight per pig per week. The material that was injected was given intraperitoneally, and a stomach tube and syringe was used to administer the material orally.

This experiment was conducted during the summer in a double-tiled insulated building. Supplemental heat was supplied with heat lamps for the first two weeks after the pigs were weaned, and then replaced with 50-watt light bulbs until the experiment was terminated.

Blood samples were obtained from the pigs in half the litters in this experiment for duplicate electrophoretic analyses of the blood serum proteins. The electrophoretic separations were run for a period of four hours at a potential of 460 volts and a current of 10 milliamps per strip. The blood samples were drawn at the end of the experiment, or three weeks after the pigs were allotted.

Table 11. Experiment 734. Summary of the gain, and the relative percent of the blood serum protein fractions

| Treatment[b] | Gain/pig[c] | Relative percent of blood serum proteins[a] | | | |
|---|---|---|---|---|---|
| | | Albumin | Alpha globulin | Beta globulin | Gamma globulin[d] |
| | (lb.) | | | | |
| Physiological saline, injected | 8.5 | 33 | 28 | 21 | 18 |
| Porcine gamma globulin, injected | 8.8 | 33 | 28 | 20 | 19 |
| Porcine gamma globulin, oral | 9.3 | 34 | 29 | 20 | 17 |
| Sow colostrum whey fraction, injected | 8.1 | 32 | 28 | 21 | 19 |
| Sow colostrum whey fraction, oral | 9.9 | 34 | 29 | 21 | 16 |

[a]Duplicate determinations on one-half the pigs.

[b]All treatments given at rate of 5 ml. per kg. of body weight per pig per week.

[c]The injection versus oral dosage of sow colostrum whey fraction and porcine gamma globulin approaches significance ($P < 0.10$).

[d]The injection versus oral dosage of sow colostrum whey fraction and porcine gamma globulin is significant ($P = 0.01$ or less).

Experiment 763

Five treatments were compared in this randomized block design experiment. Littermate groups of 5 pigs each from 15 litters were weaned and fed an average of five pounds each of prestarter ration and then the ration was changed to the starter ration shown in Table 15 in Appendix C. In this experiment the littermates from each litter were group fed. Allotment of the pigs was delayed until the first week after weaning at which time one pig from each litter was assigned to one of the five treatments. The average weight and age of the pigs when allotted was 7.2 pounds and 15.4 days, respectively. The pigs were on their experimental regime for three weeks. The treatments were administered at weekly intervals for a total of three times, namely at the beginning, the first week, and the second week the pigs were on experiment. The treatments have been shown in Table 12. The dosage rates were 5 ml. per kg. of body weight per pig, or 10 ml. per kg. of body weight per pig per week.

This experiment was conducted in the winter in a concrete block building which had a forced-air, radiant-floor-heating duct. The building temperature was maintained at and automatically heated to 70° F. A heat lamp was used in each pen as supplemental floor heat until the pigs were three weeks of age, and then a 50-watt light bulb replaced the heat lamp.

Results and discussion

Experiment 723 The results of this experiment are summarized in Table 10. The analysis of variance of these data has been shown in Table

Table 12. Experiment 763. Summary of the average gains

| Treatment[a] | Gain/pig[b] |
|---|---|
| | (lb.) |
| Physiological saline, injected, 5 ml. | 8.8 |
| Sow colostrum whey fraction, injected, 5 ml. | 6.9[c] |
| Sow colostrum whey fraction, injected, 10 ml. | 7.0 |
| Sow colostrum whey fraction, oral, 5 ml. | 9.3[c] |
| Sow colostrum whey fraction, oral, 10 ml. | 9.3[c] |

[a]Dosages administered at 5 or 10 ml. per kg. of body weight per pig per week.

[b]The injection versus oral dosage of sow colostrum whey fraction significant (P = 0.01 or less).

[c]One pig died on this treatment.

22 in Appendix D, in which the differences between treatments were no greater than might be expected for random variation. This was true for the gain per pig, feed per pound of gain, and the relative percent of gamma globulin.

Lewis (1956) has discussed the lack of response to additive factors when pigs were fed in this building. He thought it might be due to the low microorganism contamination. There was the possibility that this same phenomenon was occurring in this experiment. It appeared that the porcine gamma globulin injection had an effect on the gamma globulin level of the four pigs receiving this treatment, but due to the few numbers of pigs and the variation, this apparent higher relative percent

failed to reach significance. It appeared that each of the treatments depressed the growth of early weaned pigs.

Experiment 734 The results of this experiment have been summarized in Table 11. The statistical analysis of the data has been shown in Table 23 in Appendix D. The improved growth rate of those pigs orally dosed with either porcine gamma globulin or sow colostrum whey fraction approached significance. The injection of either of these materials increased the relative percent of gamma globulin in the blood serum. It appeared that the gamma globulin might be negatively correlated with the weight gain of the pigs in this experiment, but an analysis of covariance showed that the correlation was low and non-significant.

As would be expected, the gamma globulin percent was increased by the injection of either porcine gamma globulin solution or the sow colostrum whey fraction. On the other hand, the indication of the improved growth resulting from the oral administration of these materials was surprising. It was doubtful whether the relatively small dosage rates could account for the differences from the standpoint of nutritional supplementation.

Experiment 763 The results of this experiment have been summarized in Table 12, and the statistical analysis of the data shown in Table 24 in Appendix D. Three pigs died during this experiment. The post mortem examination revealed a herniation of the small intestine in two of the pigs and a navel infection in the other pig.

The comparison between the two methods of administration revealed that those pigs receiving the sow colostrum whey fraction orally, gained

significantly more than pigs receiving injections of whey fraction. There was no difference between the dosage rates whether injected or orally dosed. The growth depressing effect resulting from the injection of the whey fraction was not determined. Possibly an anaphylactic reaction occurred. The possibility of an infection caused by the injection was also entertained, since extreme measures in asepsis were not followed. If this had been the cause, it would seem that pigs injected with saline would not have grown as well as they did. In the previous experiment (Experiment 734), the injection of either the porcine gamma globulin or sow colostrum whey fraction did not appear to have a growth depressant effect.

## GENERAL DISCUSSION

The management practice of early weaning has been advocated for the specialized hog producer on the premise that any differences in this system compared to allowing pigs to nurse their dams for a period of eight weeks was a nutritional, housing, and management problem. It was assumed that the one-week-old pig had obtained all the passively acquired immune globulins from the colostrum and/or first milk that the pig was capable of absorbing. The report of Petersen and Campbell (1955) raised some doubt as to the soundness of this assumption. When the relative gamma globulin level of the blood serum was used as the criterion of passively acquired immunity in the baby pig, Experiment 764, there was no indication that those pigs nursing the sow beyond one week of age were absorbing additional gamma globulin. Other than the period between one and two weeks of age, the trend in the relative gamma globulin percent in the early weaned pigs and the nursing pigs was similar. This first week's difference was attributed to differences in body weight gains. This experiment also provided supplemental data on the changes in the gamma globulin level in the young pig with respect to age.

The decrease in the passively acquired immune globulins for the first few weeks after birth has been shown to occur in other species. Smith and Holm (1948) and Smith (1948) have observed this decrease in the calf. Their work indicated the half-life of the passively acquired immune globulins to be approximately 20 days. McDiarmid (1946) followed the logarithmic disappearance of agglutinins in the calf. Or-

landini et al. (1955) estimated the half-life of gamma globulin present in the human infant at birth as approximately 20 days. McCarthy and McDougall (1953) noted a decrease in agglutinins, passively acquired by lambs, which fell from high titers at birth to low values 5 to 6 weeks later. Foster et al. (1951) collected samples of blood plasma from pigs in a total of 14 litters and pooled the samples on a litter basis. Their samples showed a marked rise in the gamma globulin fraction during the first 24 hours after birth. The level of gamma globulin in the pigs showed decreases at 1 week and 3 weeks of age, with a small increase in the 8-week samples. It is possible that had samples been drawn more often in their experiment, further decreases would have been observed at 4 weeks and 5 weeks of age. The average values of gamma globulin percent found by Foster et al. (1951) were generally lower than those obtained in Experiment 764. This was undoubtedly due in part to the difference in method. Foster et al. (1951) employed the Tiselius type electrophoretic apparatus, while the paper strip electrophoretic technique was used in Experiment 764. There were several problems involved in the use of the paper strip method, especially for precise quantitative measurements, but these problems should not detract from the general gamma globulin picture nor from the comparative tests that were made in Experiment 764. Possible sources of error from the quantitative standpoint have been investigated by Franglen and Martin (1954), Klatskin et al. (1956) and Waldmann-Meyer and Schilling (1956). Dye uptake has been found to be non-linear with protein concentration. Other sources of error may be due to deviations from Beer's law in the direct

scanning of stained blood serum protein patterns on filter paper strips, and the adsorption of protein on the filter paper causing a "trailing" effect preventing complete separation of the protein fractions.

In all the studies of the disappearance and/or metabolism of passively acquired immune globulins in the young animal, there has been a source of error because the animal is growing; therefore, a dilution of the blood occurs. Thus, in order to obtain an accurate estimate of the immune globulin half-life, an accurate measurement of the blood volume would be needed.

Few studies have been designed to specifically determine the length of time after birth the newborn animal is able to absorb antibodies. The work of Halliday (1955), who used an antibody-antigen immunological technique in rats, would be considered one of the more thorough studies of this problem. Bangham and Terry (1957a) used $I^{131}$-labelled serum proteins to confirm Halliday's (1955) studies. McCarthy and McDougall (1953) were able to pin-point the ability of the lamb to absorb antibodies at somewhere between 29 and 48 hours after birth. This ability in the calf has been narrowed down to approximately the first 24 hours after birth by McAlpine and Rettger (1925), Hansen and Phillips (1947) and Henning (1953). The same was true for the newborn foal according to Bruner et al. (1950).

There was no information on the baby pig available at the time of McGirr's (1947) review. Since then, Young and Underdahl (1949) estimated the immune globulin absorptive ability in the baby pig to be limited to the first 24 hours after birth, while the estimate of Bruner

et al. (1949) was less than 2 days. The results of Experiment 778 on the study of the absorption of a specific colostral antibody from the intestinal tract showed that the 18-hour-old pigs absorbed a small amount, but the 24-hour-old pig did not. An interesting by-product of this experiment was the logarithmic linear decrease in the absorptive ability of the baby pig with respect to time. That the first order reaction rate law should hold for the ability to absorb antibodies by newborn animals may have been suspected, but it has not been shown previously.

The previous work on this problem, together with the recent histological and histochemical studies of Comline et al. (1951a, 1951b, 1953), Hill (1956), and Hill and Hardy (1956), leave little doubt that the ability to absorb antibodies by the newborn domestic animal is limited to a short period.

The results of Experiments 723, 734, and 763 cannot be fully explained. In Experiment 723 none of the treatments appeared to have a beneficial effect on the growth rate. The high molecular weight polysaccharide, Zymosan, according to Pillemer (1955), Pillemer and Ross (1955), and Pillemer et al. (1954, 1955), adsorbs the serum protein called properdin. Properdin is present in the euglobulin fraction of the blood serum, and it unites with bacteria or virus non-specifically in the immunological sense. Zymosan reduces the circulating properdin level, and this initial decline is followed by a marked increase above normal levels. Mice have shown an increased resistance to several bacterial organisms several days after intravenous administration of Zymosan. The

route of administration may have been involved in the lack of a beneficial response in early weaned pigs in Experiment 723, since administration was either per os or by intraperitoneal injection. It is suspected that the low bacterial contamination in the building in which this experiment was conducted probably had more effect on the lack of response than the method of administration.

The oral administration of porcine gamma globulin or sow colostrum whey fraction did not increase the gamma globulin level of the blood serum protein, but did appear to improve the growth rate in early weaned pigs in Experiment 734. In Experiment 763 the growth rate was improved by the oral administration of sow colostrum whey fraction. This presented an interesting situation. Since these pigs were allowed to nurse their dams for one week, there had been more than ample nursing time for the acquisition of passively acquired immunity. There was the possibility that the antibodies present in the porcine gamma globulin and sow colostrum whey fraction were effectively controlling or neutralizing some of the harmful bacteria in the lumen of the intestinal tract. The existence and effectiveness of antibodies in the lumen of the intestinal tract have been shown in cases of dysentery in humans. Harrison and Banvard (1947) and Barksdale and Ghoda (1951a, 1951b) have detected coproantibodies in the feces of patients with bacillary dysentery infections. The concentration of coproantibody level appeared to be independent of the level of the circulating blood level of the same antibody, which led to the suspicion that the source of antibodies in fecal material and in blood was not the same. This question does not appear to be

resolved. Mitchison (1953) used Thiry loop fistulas of the small intestine in rabbits in his study of active and passive antibody secretion into the loop. In his experiments the antibodies in the intestinal lumen were accounted for by leakage from the blood stream. On the other hand, Koshland (1953) concluded that fecal and serum antibodies do not have a common origin. He found that injections of Vibrio cholerae antigen produced a fecal antibody peak six days later, while the serum level rose to a maximum nine days later. The administration of the cholera vaccine plus an adjuvant resulted in the production of serum antibodies, but no fecal antibodies were detected. This he thought was due to a localized site of antibody production, and insufficient amounts of the antigen reached the site of fecal antibody production. Koshland (1953) concluded that fecal and serum antibodies do not have a common origin, and that the fecal antibodies were probably synthesized by local cells of the lymphoid-macrophage system in the lamina propia of the intestines.

It is known that the mammary gland of the domestic animal continues to secrete immune bodies through lactation, though the concentration is much lower than in colostrum. Whether the antibodies are produced in the mammary gland itself, as proposed by Campbell et al. (1957) in their "diathelic immunization" phenomenon, or whether they are produced elsewhere and transported to the mammary system via the blood stream, has not been unequivocally resolved. Regardless of where the antibodies arise, if they were effective in the lumen of the intestinal tract, the principal of "diathelic immunization" would have a bearing on the

advisability of early weaning baby pigs as a general management practice.

There were two other recent reports that relate to the question of early weaning. Hoerlein (1957) compared the immunologic response of baby pigs obtained by hysterotomy and raised in isolation to baby pigs that nursed their dams until 3 weeks of age. Four different antigens were employed in these tests. Baby pigs under 8 weeks of age which were deprived of colostrum did not produce a measurable serological response to the antigens. Pigs that had nursed their dam responded to the antigens when inoculated at 3 weeks of age. Miller et al. (1957) weaned pigs at 4 days of age, and then raised them on a complete synthetic milk diet. The antigenic response of these pigs was less than pigs that were nursing their dam when both groups were inoculated with human erythrocytes between 3 and 4 weeks of age. The explanation of these results could possibly have been determined had these investigators examined the lymphoid tissues in some of the pigs. Since the pigs were raised in isolation, it is possible that a situation similar to that reported by Reyniers et al. (1946) might account for the difference in immunological response. Reyniers et al. (1946) found that germ-free rats and chicks showed a minimal amount of quiescent lymphoid tissue, which they thought was due to a lack of immunological experience or stimuli. Further clarification on the differences, if any, between the immunological response of nursing pigs and early weaned pigs seem warranted.

# SUMMARY

Five experiments were conducted to investigate the transfer and supplementary value of passively acquired immunity in baby pigs. In one experiment four litters of 8 pigs each were divided into two equal groups at 1 week of age. Half the pigs in each litter continued to nurse their dams for an 8-week lactation, and the other pigs were weaned. Blood samples were drawn from these pigs at 1, 2, 3, 4, 5 and 8 weeks of age, and duplicate paper strip electrophoretic blood serum protein separations were made on each sample. The relative gamma globulin percent was of particular interest in evaluating whether there was any difference in passively acquired immunity in the baby pigs raised under the two above management systems. The level of relative gamma globulin percent in the early weaned pigs actually increased between 1 and 2 weeks of age and then decreased. It was suggested that this was due to the poor growth of these pigs during the initial experimental period. The relative gamma globulin percent in the nursing pigs fell normally during the period from 1 through 5 weeks of age, then showed an increase at 8 weeks of age, as did the gamma globulin concentration in the early weaned pigs after they were 2 weeks of age. The growth rate of the nursing pigs was superior to the early weaned pigs, especially during the period from 1 through 4 weeks of age. Since there were only four pigs nursing each sow, this may have been a contributing factor to the better performance of the nursing pigs. The housing was also considered as a possible factor in the poorer performance of the early weaned pigs. There was no evidence that pigs which continued to nurse their dams

beyond one week of age were acquiring immune globulins beyond this age, when the relative gamma globulin percent of their blood serum was used as the criterion of measurement.

An immunological test was employed in another experiment to obtain a precise measurement of the length of time after birth the baby pig is able to absorb immune globulins from the colostrum and/or milk secreted by the sow. High-titer colostrum was collected from 38 sows that were immunized by a series of injections with an E. coli antigen. This colostrum, which contained antibodies against a strain of E. coli, was then used to orally dose or inject the baby pigs from 16 litters. Blood samples were drawn from the baby pigs and the antibody absorption was determined by a modified sheep red blood cell hemagglutination immunologic test. It was found that the 18-hour-old baby pig was able to absorb a detectable amount of the E. coli antibody, but an insignificant amount at 24 hours of age. The ability of the baby pig to absorb the orally-ingested antibody decreased logarithmically during the period 0 to 24 hours after birth with a 50 percent decrease taking place approximately every 3 hours. It was also noted that immediately after birth the baby pig absorbed almost all the antibody ingested, when it was compared to the titers obtained when the antibody containing colostrum whey fraction was injected intraperitoneally.

Three experiments were conducted to determine the supplementary value of small amounts of porcine gamma globulin, sow colostrum whey fraction, hog cholera antiserum, and a high molecular weight polysaccharide on the relative gamma globulin level of the blood serum and

growth rate of pigs weaned at 1 week of age. In one of these experiments, the additions to the control treatment resulted in relative gamma globulin levels that were no greater than that level present in the control pigs. In a subsequent experiment, the injection of porcine gamma globulin and sow colostrum whey fraction increased the relative gamma globulin concentration of the blood serum above that present in the pigs orally dosed with these materials. However, there was an indication in this experiment that oral dosage with these two globulin-containing materials increased the growth rate of early weaned pigs to 5 weeks of age. In the last experiment of this series, two levels of orally dosed sow colostrum whey fraction increased the growth rate of early weaned pigs compared to the growth rate of comparable pigs injected with this material at two levels. It was postulated that the effect of the orally dosed whey fraction was due to its effectiveness on the harmful bacteria within the lumen of the intestinal tract.

# BIBLIOGRAPHY

Aschaffenburg, R. 1949. The nutritive value of colostrum for the calf. III. Changes in the serum protein of the newborn calf following the ingestion of small quantities of the non-fatty fraction. Brit. J. Nutrition 3:200.

_____, S. Bartlett, S. K. Kon, J. H. B. Roy, H. J. Sears, P. L. Ingram, R. Lovell, and P. C. Wood. 1952. The nutritive value of colostrum for the calf. VIII. The performance of Friesian and Shorthorn calves deprived of colostrum. J. Comp. Pathol. Therap. 62:80.

_____, _____, _____, _____, D. M. Walker, C. Briggs, and R. Lovell. 1951. The nutritive value of colostrum for the calf. V. The effect of prepartum milking. Brit. J. Nutrition 5:343.

_____, _____, _____, P. Terry, S. Y. Thompson, C. M. Walker, C. Briggs, E. Cotchin, and R. Lovell. 1949a. The nutritive value of colostrum for the calf. I. The effect of different fractions of colostrum. Brit. J. Nutrition 3:187.

_____, _____, _____, D. M. Walker, C. Briggs, E. Cotchin, and R. Lovell. 1949b. The nutritive value of colostrum for the calf. II. The effect of small quantities of the non-fatty fraction. Brit. J. Nutrition 3:196.

Askonas, B. A., P. N. Campbell, J. H. Humphrey, and T. S. Work. 1954. The source of antibody globulin in rabbit milk and goat colostrum. Biochem. J. 56:597.

Bangham, D. R. and R. J. Terry. 1957a. The absorption of $^{131}$I-labelled homologous and heterologous serum proteins fed orally to young rats. Biochem. J. 66:579.

_____ and _____. 1957b. The survival of globulins absorbed from the gut in suckling rats. Biochem. J. 66:584.

Bardelli, P. C. 1930. Sulla transmissione dell' immunità contro il tetano da madre a figlio nel cavallo. Ann. igiene. 40:675.

Barksdale, W. L. and A. Ghoda. 1951a. Agglutinating antibodies in serum and feces. J. Immunol. 66:395.

_____ and _____. 1951b. Agglutinins for Escherichia coli in serum and fecal extracts from human enteritides and from hyperimmunized and epinephrine-stimulated rabbits. J. Infectious Diseases 89:35.

Barr, M., A. T. Glenny, and K. J. Randall. 1949. Concentration of diphtheria antitoxin in cord blood and rate of loss in babies. Lancet. 257:324.

Barrick, E. R., G. Matrone, and J. C. Osborne. 1954. Effects of administering various blood serum constituents on gamma globulin levels of baby pigs. Proc. Exp. Biol. Med. 87:92.

Batty, I., F. W. R. Brambell, W. A. Hemmings, and C. L. Oakley. 1954. Selection of antitoxins by foetal membranes of rabbits. Proc. Roy. Soc. (London). B. 142:452.

Blakemore, F. 1951. Diseases of the newborn animal. I. The maternal transference of antibodies in the bovine. Vet. Record. 63:397.

_____. 1947. The maternal transference of antibodies in the bovine. Intern. Congr. Microbiol. Proc. 4:310.

_____ and R. J. Garner. 1956. The maternal transference of antibodies in the bovine. J. Comp. Pathol. Therap. 66:287.

Block, R. J., E. L. Durrum, and G. Zweig. 1955. A manual of paper chromatography and paper electrophoresis. New York, Academic Press, Inc.

Brambell, W. F. R. and R. Halliday. 1956. The route by which passive immunity is transmitted from mother to foetus in the rat. Proc. Roy. Soc. (London) B. 145:170.

_____, J. Brierley, R. Halliday, and W. A. Hemmings. 1954. Transference of passive immunity from mother to young. Lancet. 266:964.

_____, W. A. Hemmings, and M. Henderson. 1951. Antibodies and embryos. London, Athlone Press.

Briggs, C. 1951. The nutritive value of colostrum for the calf. VI. The "K" antigens of Bacterium coli. Brit. J. Nutrition 5:349.

_____, R. Lovell, R. Aschaffenburg, S. Bartlett, S. K. Kon, J. H. B. Roy, S. Y. Thompson, and D. M. Walker. 1951. The nutritive value of colostrum for the calf. VII. Observations on the nature of the protective properties of colostrum. Brit. J. Nutrition 5:356.

Bruner, D. W., R. G. Brown, F. E. Hull, and A. S. Kinkaid. 1949. Blood factors and baby pig anemia. J. Am. Vet. Med. Assoc. 115:94.

_____, E. R. Doll, F. E. Hull, and A. S. Kinkaid. 1950. Further studies on hemolytic icterus in foals. Am. J. Vet. Research 11:22.

Bulloch, W. 1898. The durability of passive diphtheria immunity. J. Path. Bacteriol. 5:274.

Campbell, B., R. M. Porter, and W. E. Petersen. 1950. Plasmacytosis of the bovine udder during colostrum secretion and experimental cessation of milking. Nature 166:913.

_____, M. Sarwar, and W. E. Petersen. 1957. Diathelic immunization--a maternal-offspring relationship involving milk antibodies. Science 125:932.

Campbell, P. N., J. H. Humphrey, and T. S. Work. 1953. The source of antibody in rabbit milk. Biochem. J. 54:XII.

Charlwood, P. A. and A. Thomson. 1948. Electrophoretic patterns of lamb serum before and after transfer of colostrum. Nature 161:59.

Cochran, W. G. and G. M. Cox. 1957. Experimental designs. 2nd ed. New York, John Wiley and Sons, Inc.

Cohen, S. G. 1950. The placental transmission of antibodies and serum $\gamma$ globulins. J. Infectious Diseases 87:291.

Comline, R. S., R. W. Pomeroy, and D. A. Titchen. 1953. Histological changes in the intestine during colostrum absorption. J. Physiol. 122:6P.

_____, H. E. Roberts, and D. A. Titchen. 1951a. Route of absorption of colostrum globulin in the newborn animal. Nature 167:561.

_____, _____, and _____. 1951b. Histological changes in the epithelium of the small intestine during protein absorption in the newborn animal. Nature 168:84.

Cooledge, L. H. 1916a. A study of the presence of Bacterium abortus (Bang) in milk. Mich. Agr. Exp. Sta. Tech. Bul. 33.

_____. 1916b. Agglutination test as a means of studying the presence of Bacterium abortus in milk. J. Agr. Research 5:871.

Culbertson, J. T. 1938. Natural transmission of immunity against Trypanosoma lewisi from mother rats to their offspring. J. Parasitol. 24:65.

_____. 1939a. The immunization of rats of different age groups against Trypanosoma lewisi by the administration of specific antiserum per os. J. Parasitol. 25:181.

_____. 1939b. Transmission of resistance against Trypanosoma lewisi from a passively immunized mother rat to young nursing upon her. J. Parasitol. 25:182.

Deutsch, H. F. 1947. A study of whey proteins from the milk of various animals. J. Biol. Chem. 169:437.

Earle, I. P. 1935. Influence of the ingestion of colostrum on the proteins of the blood sera of young foals, kids, lambs, and pigs. J. Agr. Research 51:479.

Ehrlich, P. 1892. Ueber immunität durch vererbung und säugung. Z. Hyg. Infektionskrankh. 12:183.

Famulener, L. W. 1912. On the transmission of immunity from mother to offspring. A study upon serum hemolysins in goats. J. Infectious Diseases 10:332.

Filmer, D. B. and T. J. McClure. 1951. Absorption of anti-nematode antibodies from ewe's colostrum by the new-born lamb. Nature 168: 170.

Foster, J. F., R. W. Friedell, D. Catron, and M. R. Dieckmann. 1951. Electrophoretic studies on swine. III. Composition of baby pig plasma and sow's whey during lactation. Arch. Biochem. Biophys. 31: 104.

Franglen, G. T. and N. H. Martin. 1954. The interaction of dyes with proteins on paper with special reference to electrophoresis. Biochem. J. 57:626.

Friedell, R. W., J. F. Foster, D. Catron, and M. R. Dieckmann. 1951. Electrophoretic studies on swine. II. The composition of plasma during gestation and lactation. Iowa State College J. Science 25: 521.

Gruskay, F. L. and R. E. Cooke. 1955. The gastrointestinal absorption of unaltered protein in normal infants and in infants recovering from diarrhea. Pediatrics 16:763.

Halliday, R. 1955. The absorption of antibodies from immune sera by the gut of the young rat. Proc. Roy. Soc. (London) B. 143:408.

_____. 1956. The termination of the capacity of young rats to absorb antibody from the milk. Proc. Roy. Soc. (London) B. 145:179.

Hansen, R. G. and P. H. Phillips. 1947. Studies on proteins from bovine colostrum. I. Electrophoretic studies on the blood serum proteins of colostrum-free calves and of calves fed colostrum at various ages. J. Biol. Chem. 171:223.

_____ and _____. 1949. Studies on proteins from bovine colostrum. III. The homologous and heterologous transfer of ingested protein to the blood stream of the young animal. J. Biol. Chem. 179:523.

Harris, T. N. and S. Harris. 1956. The genesis of antibodies. Am. J. Med. 20:114.

Harrison, P. E. and J. Banvard. 1947. Coproantibody excretion during enteric infections. Science 106:188.

Hartley, G., Jr. 1942. The permeability of the gastrointestinal mucosa of guinea pigs to crystalline egg-albumin. J. Immunol. 43:297.

Hartley, P. 1951. The effect of peptic digestion on the properties of diphtheria antitoxin. Proc. Roy. Soc. (London) B. 138:499.

Hayes, F. 1921. Some studies in swine abortion. J. Am. Vet. Med. Assoc. 60:435.

Heidelberger, M., H. P. Treffers, R. Schoenheimer, S. Ratner, and D. Rittenberg. 1942. Behavior of antibody protein toward dietary nitrogen in active and passive immunity. J. Biol. Chem. 144:555.

Henning, M. W. 1953. Calf paratyphoid. III. The transmission of antibodies to newly-born calves. Onderstepoort J. Vet. Research 26:45.

Hill, K. J. 1956. Gastric development and antibody transference in the lamb, with some observations on the rat and guinea pig. Quart. J. Exp. Physiol. 41:421.

_____ and W. S. Hardy. 1956. Histological and histochemical observations on the intestinal cells of lambs and kids absorbing colostrum. Nature 178:1353.

Hoerlein, A. B. 1952. Studies in swine brucellosis. I. The pathogenesis of artificial Brucella melitensis infection. Am. J. Vet. Research 13:67.

_____. 1957. The influence of colostrum on antibody response in baby pigs. J. Immunol. 78:112.

_____, C. H. Adams, and R. J. Meade. 1956. Hysterotomy to obtain "disease free" baby pigs. J. Am. Vet. Med. Assoc. 128:127.

Howard, F. A. and M. I. T. Cronin. 1955. Colostral transfer of anti-erythrocyte agglutinins from mare to foal. J. Am. Vet. Med. Assoc. 126:93.

Howe, P. E. 1921. An effect of the ingestion of colostrum upon the composition of the blood of new-born calves. J. Biol. Chem. 49: 115.

_____. 1922. The differential precipitation of the proteins of colostrum and a method for the determination of the proteins in colostrum. J. Biol. Chem. 52:51.

Jameson, E., C. Alvarez-Tostado, and H. H. Sortor. 1942. Electrophoretic studies on new-born calf serum. Proc. Soc. Exp. Biol. Med. 51:163.

Kastelic, J., O. G. Bentley, and P. H. Phillips. 1950. Studies on growth and survival of calves fed semi-synthetic milks from birth. J. Dairy Sci. 33:725.

Klatskin, G., O. M. Reinmuth, and W. Barnes. 1956. A study of the densitometric method of analyzing filter paper. J. Lab. Clin. Med. 48:476.

Kolmer, J. A. and F. Boerner. 1945. Approved laboratory technic. 4th ed. New York, D. Appleton-Century Co.

Kolouch, F., R. A. Good, and B. Campbell. 1947. The reticuloendothelial origin of the bone marrow plasma cells in hypersensitive states. J. Lab. Clin. Med. 32:749.

Koshland, M. E. 1953. The origin of fecal antibody and its relationship to immunization with adjuvant. J. Immunol. 70:359.

Larson, B. L. and K. A. Kendall. 1957. Changes in specific blood serum protein levels associated with parturition in the bovine. J. Dairy Sci. 40:659.

Laskowski, M., Jr. and M. Laskowski. 1951. Crystalline trypsin inhibitor from colostrum. J. Biol. Chem. 190:563.

_____, P. H. Mars, and M. Laskowski. 1952. Comparison of trypsin inhibitor from colostrum with other crystalline trypsin inhibitors. J. Biol. Chem. 198:745.

Lemétayer, E., L. Nicol, L. Jacob, O. Girard, and R. Corvazier. 1946. Immunité antitoxique diaplacentaire du poulain issu de juments immunisées. Compt. Rend. Soc. Biol. 140:852.

Lewis, C. J. 1956. Qualitative and quantitative studies on proteolytic digestive enzymes in baby pig nutrition. Unpublished Ph. D. Thesis. Ames, Iowa, Iowa State College Library.

Lewis, J. H. and H. G. Wells. 1922. The function of the colostrum. J. Am. Med. Assoc. 78:863.

Lippard, V. W., O. M. Schloss, and P. A. Johnson. 1936. Immune reactions induced in infants by intestinal absorption of incompletely digested cow's milk protein. Am. J. Diseases Children 51:562.

Little, R. B. and M. L. Orcutt. 1922. The transmission of Bacillus abortus from cow to calf in the colostrum. J. Exp. Med. 35:161.

Mason, J. H., T. Dalling, and W. S. Gordon. 1930. Transmission of maternal immunity. J. Path. Bacteriol. 33:783.

McAlpine, J. G. and L. F. Rettger. 1925. Serological studies on bovine infectious abortion. J. Immunol. 10:811.

McCarthy, E. F. and E. I. McDougall. 1949. Absorption of immune globulin by the young lamb after ingestion of colostrum. Nature 164: 354.

_____ and _____. 1953. Absorption of immune globulin by the young lamb after ingestion of colostrum. Biochem. J. 55:177.

McDiarmid, A. 1946. The transference of agglutinins for Brucella abortus from cow to calf and their persistence in the calf's blood. Vet. Record 58:146.

McGirr, J. L. 1947. Colostral transmission of antibody substances from mother to offspring. Vet. J. 103:345.

Miller, E. R., D. A. Schmidt, D. E. Ullrey, J. A. Hoefer, and R. W. Luecke. 1957. Differences in antibody production between nursing pigs and those fed synthetic milk diets. J. Ani. Sci. 16: In press.

Mitchison, N. A. 1953. The passage of antibodies into the intestine in rabbits. Quart. J. Exp. Physiol. 38:139.

Nelson, J. B. 1932. The maternal transmission of vaccinial immunity in swine. J. Exp. Med. 56:835.

Nelson, L. F. 1953. Development of practical synthetic milk formulas for baby pigs. Unpublished M. S. Thesis. Ames, Iowa, Iowa State College Library.

Neter, E., E. A. Gorzynski, R. M. Gino, O. Westphal, and O. Lüderitz. 1956. The enterobacterial hemagglutination test and its diagnostic potentialities. Can. J. Microbiol. 2:232.

Orcutt, M. L. and P. E. Howe. 1922. The relation between the accumulation of globulins and the appearance of agglutinins in the blood of new-born calves. J. Exp. Med. 36:291.

Orlandini, O., A. Sass-Kortsak, and J. H. Ebbs. 1955. Serum gamma globulin levels in normal infants. Pediatrics 16:575.

Oxer, D. T. 1936. The transmission of antitoxic immunity from the ewe, vaccinated against entero-toxaemia to the lamb. Australian Vet. J. 12:54.

Petersen, W. E. and B. Campbell. 1955. Use of protective principles in milk and colostrum in prevention of disease in man and animals. J. Lancet. 75:494.

Pillemer, L. 1955. The properdin system. Trans. N. Y. Acad. Sci. 17: 526.

_____, L. Blum, I. H. Lepow, O. A. Ross, E. W. Todd, and A. C. Wardlaw. 1954. The properdin system and immunity. I. Demonstration and isolation of a new serum protein, properdin, and its role in immune phenomena. Science 120:279.

_____ and O. A. Ross. 1955. Alterations in serum properdin levels following injections of zymosan. Science 121:732.

_____, M. D. Schoenberg, L. Blum, and L. Wurz. 1955. Properdin system and immunity. II. Interaction of the properdin system with polysaccharides. Science 122:545.

Polson, A. 1943. Variation of serum composition with the age of horses as shown by electrophoresis. Nature 152:413.

_____. 1952. Comparative electrophoretic studies of bovine and human colostrum in relation to neo-natal immunity. Onderstepoort J. Vet. Research 25, no.4:7.

Ratner, B. and H. L. Gruehl. 1934. Passage of native proteins through the normal gastro-intestinal wall. J. Clin. Invest. 13:517.

Raymond, S. 1955. Paper electrophoresis. 3rd ed. New York, E-C Apparatus Co.

Reymann, G. C. 1920. On the transfer of the so-called normal-antibodies from mother to offspring. J. Immunol. 5:227.

Reyniers, J. A., P. C. Trexler, and R. F. Ervin. 1946. Rearing germ-free albino rats. Lobund Reports. Laboratory of Bacteriology, Notre Dame University. no. 1.

Roberts, H. E., A. N. Worden, and E. T. Rees Evans. 1954. Observations on some effects of colostrum deprivation in the calf. J. Comp. Pathol. Therap. 64:283.

Roy, J. H. B., J. Palmer, K. W. G. Shillam, P. L. Ingram, and P. C. Wood. 1955a. The nutritive value of colostrum for the calf. X. The relationship between the period of time that a calfhouse has been occupied and the incidence of scouring and mortality in young calves. Brit. J. Nutrition 9:11.

_____, K. W. G. Shillam, J. Palmer, and P. L. Ingram. 1955b. The nutritive value of colostrum for the calf. XI. The effect of aureomycin on the performance of colostrum-deprived calves. Brit. J. Nutrition 9:94.

San Clemente, C. L. and I. F. Huddleson. 1943. Electrophoretic studies of the proteins of bovine serums with respect to Brucella. Mich. Agr. Exp. Sta. Tech. Bul. 182.

Schneider, L. and J. Szathmáry. 1938. Ueber die immunität der neugeborenen säugetiere. Z. Immunitätsforsch. 94:458.

_____ and _____. 1939. Ueber die immunität der neugeborenen säugetiere. Z. Immunitätsforsch. 95:465.

Schoenheimer, R., S. Ratner, and D. Rittenberg. 1942. The interaction of antibody protein with dietary nitrogen in actively immunized animals. J. Biol. Chem. 144:545.

Shuman, R. D., F. L. Earl, and J. W. Stevenson. 1956. Atrophic rhinitis. VI. The establishment of an atrophic rhinitis-free herd of hogs. J. Am. Vet. Med. Assoc. 128:189.

Smith, E. L. 1946. The immune proteins of bovine colostrum and plasma. J. Biol. Chem. 164:345.

_____. 1948. The isolation and properties of the immune proteins of bovine milk and colostrum and their role in immunity: a review. J. Dairy Sci. 31:127.

_____ and N. H. Coy. 1946. The absorption spectra of immune proteins. J. Biol. Chem. 164:367.

_____ and R. D. Greene. 1947. Further studies on the amino acid composition of immune proteins. J. Biol. Chem. 171:355.

_____, _____, and E. Bartner. 1946. Amino acid and carbohydrate analyses of some immune proteins. J. Biol. Chem. 164:359.

_____ and A. Holm. 1948. The transfer of immunity to the new-born calf from colostrum. J. Biol. Chem. 175:349.

Smith, T. 1925. Hydropic stages in the intestinal epithelium of newborn calves. J. Exp. Med. 41:81.

_____. 1930. The immunological significance of colostrum. I. The relation between colostrum, serum, and the milk of cows normal and immunized towards B. coli. J. Exp. Med. 51:473.

_____ and R. B. Little. 1922. The significance of colostrum to the newborn calf. J. Exp. Med. 36:181.

_____ and _____. 1930. The immunological significance of colostrum. II. The initial feeding of serum from normal cows and cows immunized towards B. coli in place of colostrum. J. Exp. Med. 51: 483.

_____ and M. L. Orcutt. 1925. The bacteriology of the intestinal tract of young calves with special reference to the early diarrhea ("scours"). J. Exp. Med. 41:89.

Snedecor, G. W. 1956. Statistical methods. 5th ed. Ames, Iowa, Iowa State College Press.

Speer, V., G. Ashton, F. Diaz, and D. Catron. 1954. New I.S.C. prestarter "75". Iowa Farm Sci. 8, no. 10:3.

Waldmann-Meyer, H. and K. Schilling. 1956. Protein adsorption on filter paper. Science 124:1028.

Walzer, M. 1927. Studies in absorption of undigested proteins in human beings. I. A simple direct method of studying the absorption of undigested protein. J. Immunol. 14:143.

Westphal, O., O. Lüderitz, and F. Bister. 1952a. Über die extraktion von bakterien mit phenol/wasser. Z. Naturforsch. 7b:148.

_____, _____, E. Eichenberger, and W. Keiderling. 1952b. Über bakterielle reizstoffe. I. Mitt: Reindarstellung eines polysaccharid-pyrogens aus Bacterium coli. Z. Naturforsch. 7b:536.

Whitehair, C. K. and C. M. Thompson. 1956. Observations on raising "disease free" swine. J. Am. Vet. Med. Assoc. 128:94.

Wiener, A. S. 1951. The half-life of passively acquired antibody globulin molecules in infants. J. Exp. Med. 94:213.

Wisconsin Agricultural Experiment Station. 1957. Seventy-third annual report for year ending June 30, 1956. Pt. 1:49.

Young, G. A. and N. R. Underdahl. 1953. Isolation units for growing baby pigs without colostrum. Am. J. Vet.Research 14:571.

_____, _____, and R. W. Hinz. 1955. Procurement of baby pigs by hysterectomy. Am. J. Vet. Research 16:123.

Young, G. A., Jr. and N. R. Underdahl. 1949. Swine influenza as a possible factor in suckling pig mortalities. II. Colostral transfer of hemagglutinin inhibitors for swine influenza virus from dam to offspring. Cornell Vet. 39:120.

_____ and _____. 1950. Neutralization and hemagglutination inhibition of swine influenza virus by serum from suckling swine and by milk from their dams. J. Immunol. 65:369.

ACKNOWLEDGMENTS

The author wishes to express his indebtedness to Dr. Damon V. Catron for his invaluable guidance, counsel and encouragement throughout the author's graduate program. Acknowledgment is also extended to Dr. Loyd Y. Quinn and Dr. R. S. Allen for direction and provision of facilities needed to carry out portions of the experimental work reported herein. Appreciation is extended to Herbert Brown and Don Quinn for their assistance in the collection of the data, and to Paul G. Homeyer for his assistance and counsel in the statistical analyses of the data.

# APPENDIX A

A barbital buffer with a pH of 8.6 and ionic strength of 0.1 was used in all separations. The buffer was made up in 5 liter batches using 14 grams of diethyl barbituric acid and 103 grams of sodium barbital.

Each of the three compartments of the A-C Electrophoretic Apparatus was filled to within one to two mm. of the top, and adjustment between the cells was made by opening the leveling tubes. Whatman No. 3 MM filter paper strips 2.5 inches by 18.5 inches were used in all runs. Six strips could be run simultaneously. Before the samples were applied to the strips they were moistened with the buffer. Three dry strips were placed on the cooling plate with the ends extending over the end walls of the buffer cells. Each strip was moistened with 10 ml. of buffer as evenly as possible, and then three dry strips were placed on each of the moist strips. The lid was clamped in place for approximately 10 minutes to obtain even distribution of the buffer. Three samples of whole blood serum were then applied to each strip by using a straight edge as a guide and a micropipette which delivered approximately 0.01 ml. of serum in a band approximately 1 cm. wide and 9 cm. from the center of the strip. A solution of 0.9 percent sodium chloride was used to rinse the pipette between samples.

The separations were carried cut with a potential of 460 volts and a current of 10 milliamps per strip in some experiments and 150 volts and 2.5 milliamps per strip in some experiments. Water with a temperature of approximately 15° C. ran through the cooling plates continuously.

At the completion of the separations, the strips were placed in a 110° C. drying oven for 30 minutes. The strips were then placed in a dye bath for 16 to 24 hours. The staining solution included 0.1 gram bromphenol blue, 50 grams zinc sulfate, and 50 ml. glacial acetic acid diluted to 1 liter. After being stained, the patterns were bathed in three rinses of 2 percent acetic acid solution for periods of 5, 5 and 10 minutes, respectively, and then in one rinse of 2 percent sodium acetate and 10 percent acetic acid solution for 2 minutes. The strips were then placed in the oven to dry at 110° C.

Prior to scanning, the patterns were placed in a mineral oil bath, suspended vertically to allow the oil to drain off, and then the excess oil was removed by blotting the strips between pieces of filter paper. Optical density readings were then obtained by running the sample through a Photovolt Corporation, New York, Model No. 501A photometer and Model No. 52-C scanner. Graphs were plotted directly on graph paper placed on the movable table mounting attached to the spectrophotometric scanner. Readings were recorded every 2 mm.

Area measurements for the various fractions were obtained with a Keuffel and Esser Company, New York, 4236M planimeter, and then the values were converted to relative percent.

# APPENDIX B

A strain of Escherichia coli isolated from the cecum of a rabbit was used as the source of antigen in all the hemagglutination tests in one phase of the study reported herein. This organism was agglutinated by E. coli type O111:B4 antisera obtained from the American Type Culture Collection, Washington 7, D. C. The latter strain has been associated with numerous outbreaks of epidemic diarrhea in young infants (Neter et al., 1956).

In the extraction of the lipopolysaccharide fraction by the method of Westphal et al. (1952a, 1952b), 10 liters of nutrient broth were sterilized and inoculated with the E. coli strain isolated from a rabbit. After 24 hours growth with aeration at 37° C., the medium was centrifuged in a Sharples centrifuge and the organisms were collected. Ten grams of acetone dried bacteria was then suspended in 160 ml. of distilled water and cooled to 2° C., and 265 ml. of 75 percent phenol was then added slowly with agitation. After standing 30 minutes in the cold with occasional shaking, the mixture was centrifuged in the cold (1500 r.p.m. for 30 minutes), the supernatant decanted, and the cells discarded. The phenol-water extract containing the lipopolysaccharide antigenic substances was then dialyzed for 2 days against tap water and then for 1 day against distilled water. The material was filtered, 6 volumes of ethyl alcohol added, 10 ml. of ethyl alcohol saturated with sodium acetate added, and was again filtered. The precipitate was dried in a vacuum oven, and after being ground, it was used in the preparation of the modified sheep red blood cells.

Sheep blood was collected using sodium citrate anticoagulant, and was then centrifuged. The plasma was discarded, and the red blood cells were washed three times in 0.85 percent sodium chloride. The red cells were then resuspended in 0.85 percent saline to give a 2.5 percent red cell concentration. The lipopolysaccharide antigen was suspended in a small amount of 0.85 percent saline and heated to 100° C. for one hour. After being cooled, the lipopolysaccharide solution was then added to the red cells at the rate of 1.5 mg. per ml. of red cell suspension. The mixture of red cells and lipopolysaccharide was incubated at 37° C. for 30 minutes, the mixture was then centrifuged, and the modified cells washed three times with saline to remove the excess lipopolysaccharide. The modified red cells were again resuspended in 0.85 percent saline with a 2.5 percent red cell concentration.

Before the serum samples were tested for specific antibody activity, the samples were inactivated for 30 minutes in a 56° C. water bath. Twofold serial dilutions were then carried out in small test tubes by adding 0.1 ml. of serum to the first tube, which contained 0.4 ml. of 0.85 percent saline, and then transferring 0.25 ml. from the first tube to the subsequent tube, which contained 0.25 ml. saline. Then 0.25 ml. of the 2.5 percent modified red cell solution was added to each tube. The tubes were then incubated at 37° C. for 8 hours, and then read grossly by gently tapping each tube and noting whether the red cells were clumped. The same procedure was used in testing the colostrum samples.

The standardized antigen was prepared by growing the E. coli organism on agar for 24 hours at 37° C. The cell growth was then rinsed off the agar and the suspension centrifuged in a Sharples centrifuge, and the organisms collected. After being washed, they were suspended in a solution of 0.85 percent saline containing 0.2 percent formalin. Enough cells were added to this suspension to give a reading of 3.0 on the McFarland Nephalometer, or approximately 900 million cells per ml., according to the method of Kolmer and Boerner (1945).

The sow colostrum whey fraction was separated by adding rennin to the whole colostrum. Cheese rennet tablets obtained from the Charles Hansen's Laboratory, Incorporated, Little Falls, New York were added at the rate of one-fourth tablet per 1000 ml. of colostrum. The tablet was first dissolved in a small amount of distilled water then added to colostrum, which had been previously warmed to 30° C. The solution was stirred vigorously for one minute then allowed to stand for approximately 30 minutes until a firm curd had formed. The material was then centrifuged at 2000 r.p.m. and the whey fraction decanted and filtered through sterile cheesecloth.

# APPENDIX C

Table 13. Composition of the prestarter ration fed in Experiments 764, 723, 734 and 763

| Ingredients[a] | Amounts |
|---|---|
| Cane sugar | 10.00 |
| Corn sugar | 5.00 |
| Ground yellow corn | 14.70 |
| Ground oat groats | 5.00 |
| Dried whey (sweet) | 10.00 |
| Dried skimmilk (low heat, spray dried) | 40.00 |
| Solvent soybean oil meal (50% protein) | 0.50 |
| Fish solubles (condensed) | 2.50 |
| Corn steep water | 1.00 |
| Dried brewers' yeast | 1.00 |
| Dried beet pulp | 2.00 |
| Stabilized lard | 5.00 |
| Dicalcium phosphate | 0.90 |
| Iodized salt | 0.25 |
| Trace mineral mix[b] | 0.15 |
| Vitamin premix (soybean meal carrier) | 2.00 |
| Total (lbs.) | 100.00 |

[a]The calculated analysis of this ration is shown in Table 19.

[b]Composition of trace mineral mix is shown in Table 18.

Table 14. Composition of the starter ration fed in Experiments 723 and 734

| Ingredients[a] | Amounts |
|---|---|
| Cane sugar | 15.00 |
| Ground yellow corn | 31.10 |
| Ground oat groats | 20.00 |
| Dried whey (sweet) | 2.50 |
| Dried skimmilk (low heat, spray dried) | 10.00 |
| Solvent soybean oil meal (44% protein) | 14.00 |
| Fish solubles (condensed) | 2.50 |
| Calcium carbonate | 0.45 |
| Dicalcium phosphate | 1.80 |
| Iodized salt | 0.50 |
| Trace mineral mix[b] | 0.15 |
| Vitamin premix (soybean meal carrier) | 2.00 |
| Total (lbs.) | 100.00 |

[a]The calculated analysis of this ration is shown in Table 19.

[b]Composition of trace mineral mix is shown in Table 18.

Table 15. Composition of starter ration fed in Experiments 763 and 764

| Ingredients[a] | Amounts |
|---|---|
| Cane sugar | 15.00 |
| Ground yellow corn | 30.25 |
| Ground oat groats | 20.00 |
| Solvent soybean oil meal (50% protein) | 13.25 |
| Dried skimmilk (low heat, spray dried) | 10.00 |
| Dried whey (sweet) | 2.50 |
| Fish solubles (condensed) | 2.00 |
| Stabilized lard | 2.00 |
| Calcium carbonate | 0.55 |
| Dicalcium phosphate | 1.80 |
| Iodized salt | 0.50 |
| Trace mineral mix[b] | 0.15 |
| Vitamin premix (soybean meal carrier) | 2.00 |
| Total (lbs.) | 100.00 |

[a]The calculated analysis of this ration is shown in Table 19.

[b]Composition of trace mineral mix is shown in Table 18.

Table 16. Composition of the grower ration fed in Experiment 764

| Ingredients[a] | Amounts |
|---|---|
| Ground yellow corn | 75.90 |
| Solvent soybean oil meal (50% protein) | 14.75 |
| Meat and bone scraps (50% protein) | 2.50 |
| Dried whey (sweet) | 2.50 |
| Calcium carbonate | 0.30 |
| Dicalcium phosphate | 1.40 |
| Iodized salt | 0.50 |
| Trace mineral mix[b] | 0.15 |
| Vitamin premix (soybean meal carrier) | 2.00 |
| Total (lbs.) | 100.00 |

[a]The calculated analysis of this ration is shown in Table 19.

[b]Composition of trace mineral mix is shown in Table 18.

Table 17. Composition of the lactation ration fed to the sows in Experiments 764 and 778

| Ingredients[a] | Amounts |
|---|---|
| Ground yellow corn | 40.00 |
| Ground oats | 20.00 |
| Dried beet pulp | 5.00 |
| Solvent soybean oil meal (50% protein) | 10.00 |
| Meat and bone scraps (50% protein) | 5.00 |
| Dried whey (sweet) | 5.00 |
| Dehydrated alfalfa meal | 10.00 |
| Fish solubles (condensed) | 2.50 |
| Dicalcium phosphate | 0.80 |
| Iodized salt | 0.50 |
| Trace mineral mix[b] | 0.20 |
| Vitamin - antibiotic premix (corn carrier) | 1.00 |
| Total (lbs.) | 100.00 |

[a]The calculated analysis of this ration is shown in Table 19.

[b]Composition of trace mineral mix is shown in Table 18.

Table 18. Contents of trace mineral mix used in the rations of Experiments 764, 778, 723, 734 and 763

| Element | Amounts in parts per million | |
|---|---|---|
| | Contributed to diet when added at 0.15% | Contributed to diet when added at 0.20% |
| Iron | 105.6 | 140.8 |
| Copper | 7.2 | 9.6 |
| Cobalt | 2.4 | 3.2 |
| Zinc | 122.4 | 163.2 |
| Manganese | 85.2 | 113.6 |
| Potassium | 11.2 | 15.0 |

Table 19. Calculated analyses of the rations fed in Experiments 764, 778, 723, 734, and 763

| Item[a] | Prestarter (Table 13) | Starter (Table 14) | Starter (Table 15) | Grower (Table 16) | Lactation (Table 17) |
|---|---|---|---|---|---|
| Protein, % | 20.10 | 18.00 | 18.00 | 16.20 | 16.80 |
| Calcium, % | 0.87 | 0.85 | 0.86 | 0.80 | 0.98 |
| Phosphorus, % | 0.71 | 0.70 | 0.70 | 0.71 | 0.70 |
| Vitamin A, I.U. | 10,147.0 | 3011.0 | 3003.0 | 2000.0 | 7900.0 |
| Vitamin $D_2$, I.U. | 1000.0 | 500.0 | 500.0 | 400.0 | 500.0 |
| Riboflavin, mg. | 5.0 | 4.2 | 4.0 | 3.0 | 2.6 |
| Calcium pantothenate, mg. | 10.0 | 8.1 | 8.0 | 6.0 | 9.0 |
| Niacin, mg. | 29.7 | 25.2 | 25.0 | 20.0 | 20.0 |
| Choline, mg. | 504.0 | 457.0 | 456.0 | 400.0 | 432.0 |
| Vitamin $B_{12}$, mcg.[b] | 20.0 | 20.0 | 20.0 | 10.0 | 10.0 |
| Thiamine, mg. | 2.0 | -- | -- | -- | -- |
| Pyridoxine, mg. | 2.0 | -- | -- | -- | -- |
| Para-amino benzoic acid, mg. | 8.0 | -- | -- | -- | -- |
| Vitamin K, mg. | 1.0 | -- | -- | -- | -- |
| Vitamin E, mg. | 10.0 | -- | -- | -- | -- |
| Folic acid, mcg. | 9.0 | -- | -- | -- | -- |
| Ascorbic acid, mg. | 300.0 | -- | -- | -- | -- |
| Penicillin, mg. | -- | -- | -- | -- | 5.0 |

[a]The vitamins and antibiotic are listed as amounts per pound of ration.

[b]The vitamins and antibiotic listed from vitamin $B_{12}$ and below are the amounts added per pound of ration and do not include that present in the natural ingredients.

Table 20. Experiment 764. Analysis of variance plan and mean squares for gain, albumin, alpha globulin, beta globulin, and gamma globulin

| Source of variation | Degrees of freedom | Mean squares Gain | Degrees of freedom | Mean squares Albumin | Alpha globulin | Beta globulin | Gamma globulin |
|---|---|---|---|---|---|---|---|
| Litters | 3 | 21.93[a] | 3 | 118.32[b] | 295.01[b] | 118.09[a] | 1315.27[b] |
| Pairs/litters | 12 | 9.08 | 12 | 22.00 | 9.38 | 8.25 | 42.36 |
| Treatments | 1 | 39.60[b] | 1 | 1063.34[b] | 251.88[b] | 21.56 | 455.01[a] |
| Treatments x litters | 3 | 1.60 | 3 | 7.65 | 9.13 | 41.71 | 91.17 |
| Pairs x treatments/litters | 12 | 3.25 | 12 | 19.70 | 15.11 | 22.47 | 54.34 |
| Weeks | 4 | 2132.78[b] | 5 | 668.80[b] | 13.48 | 85.78[b] | 550.19[b] |
| Weeks x litters | 12 | 9.86[a] | 15 | 29.41[b] | 17.33[b] | 10.53 | 31.13[a] |
| Weeks x pairs/litters | 48 | 3.83 | 60 | 8.33 | 7.24 | 6.33 | 13.93 |
| Weeks x treatments | 4 | 27.44[b] | 5 | 179.55[b] | 50.83[b] | 37.77[b] | 142.45[b] |
| Weeks x treatments x litters | 12 | 1.67 | 15 | 5.99 | 5.50 | 12.39 | 14.48 |
| Weeks x treatments x pairs/litters | 48 | 1.07 | 60 | 8.71 | 6.42 | 7.06 | 12.86 |
| Determinations | -- | | 192 | 5.15 | 2.59 | 4.24 | 3.56 |

[a]Significant at P = 0.05 or less.

[b]Significant at P = 0.01 or less.

Table 21. Experiment 778 (Group III). Analysis of covariance of the hemagglutination titers with respect to hours after birth

| Source of variation | Degrees of freedom | Mean square |
|---|---|---|
| Litters | 7 | 0.31 |
| Treatment (hour of dosage) | 4 | 7.20[a] |
| Linear regression $\frac{(\Sigma xy)^2}{\Sigma x^2}$ | 1 | 27.92[a] |
| Deviations from regression | 3 | 0.29 |
| Error | 28 | 0.17 |

[a]Significant at P = 0.01 or less.

Table 22. Experiment 723. Analysis of variance plan and mean squares for gain, feed and serum gamma globulin

| Source of variation | Degrees of freedom | Mean squares | | |
|---|---|---|---|---|
| | | Gain | Feed/lb. gain | Gamma globulin |
| Replications | 3 | 23.00 | 0.0898 | 5.41 |
| Treatments | 8 | 14.83 | 0.0638 | 7.90 |
| Blocks (adjusted) | 8 | 12.46 | 0.0528 | 1.56 |
| Intra-block | 16 | 7.73 | 0.0943 | 3.64 |

Table 23. Experiment 734. Analysis of variance plan and mean squares for gain and serum gamma globulin

| Source of variation | Degrees of freedom | Mean square Gain | Degrees of freedom | Mean square Gamma globulin |
|---|---|---|---|---|
| Replications (litters) | 19 | 17.63 | 9 | 63.20[a] |
| Treatments | 4 | 10.05 | 4 | 34.80[b] |
| Saline vs. others | 1 | 4.30 | 1 | 2.60 |
| Gamma globulin vs. colostrum | 1 | .00 | 1 | 9.80 |
| Injection vs. oral dosage | 1 | 27.50 | 1 | 125.00[a] |
| Remainder | 1 | 8.38 | 1 | 1.60 |
| Replication x treatment | 76 | 7.31 | 36 | 9.30[b] |
| Determination within sample | | | 50 | 3.60 |

[a]Significant at P = 0.01 or less.

[b]Significant at P = 0.05 or less.

Table 24. Experiment 763. Analysis of variance plan and mean squares for gain

| Source of variation | Degrees of freedom | Mean square Gain |
|---|---|---|
| Replications (litters) | 14 | 6.94 |
| Treatments | 4 | 27.65[a] |
| Saline vs. others | 1 | 3.88 |
| 1X level vs. 2X level | 1 | 1.15 |
| Injection vs. oral dosage | 1 | 103.75[a] |
| Remainder | 1 | 1.84 |
| Replications x treatments | 53 | 5.83 |

[a]Significant at P = 0.01 or less.

Table 25. Experiment 778 (sows). Dosage schedule and rate of injection of E. coli antigen in sows for the production of high-titer colostrum[a]

| Group | Injection no. | 1 | 2 | 3 | 4 | 5 | 6 | 7 |
|---|---|---|---|---|---|---|---|---|
| I | Route of injection[b] | Subcut. | Subcut. | Subcut. | I.V. | I.V. | I.V. | I.V. |
| | Rate (ml.) | 2.0 | 4.0 | 8.0 | 0.5 | 3.0 | 3.0 | 3.0 |
| | No. days after initial inj. | 0 | 7 | 14 | 19 | 23 | 30 | 37 |
| | No. sows farrowed after injection no. | - | - | - | - | 3 | 5 | 1 |
| II | Route of injection | Subcut.&I.V. | I.V. | I.V. | I.V. | I.V. | I.V. | I.V. |
| | Rate (ml.) | 0.5 ea. | 2.0 | 3.0 | 4.0 | 4.0 | 4.0 | 4.0 |
| | No. days after initial inj. | 0 | 4 | 9 | 16 | 23 | 30 | 37 |
| | No. sows farrowed after injection no. | - | - | - | 3 | 8 | 5 | 1 |
| III | Route of injection | I.V. | I.V. | I.V. | I.V. | I.V. | I.V. | I.V. |
| | Rate (ml.) | 2.0 | 4.0 | 5.0 | 5.0 | 5.0 | 5.0 | 5.0 |
| | No. days after initial inj. | 0 | 5 | 12 | 19 | 26 | 33 | 40 |
| | No. sows farrowed after injection no. | - | - | - | - | 5 | 5 | 2 |

[a]The resulting colostrum titers on these sows are shown in Figures 7, 8 and 9.

[b]Subcut. and I.V. are subcutaneous and intravenous injections.

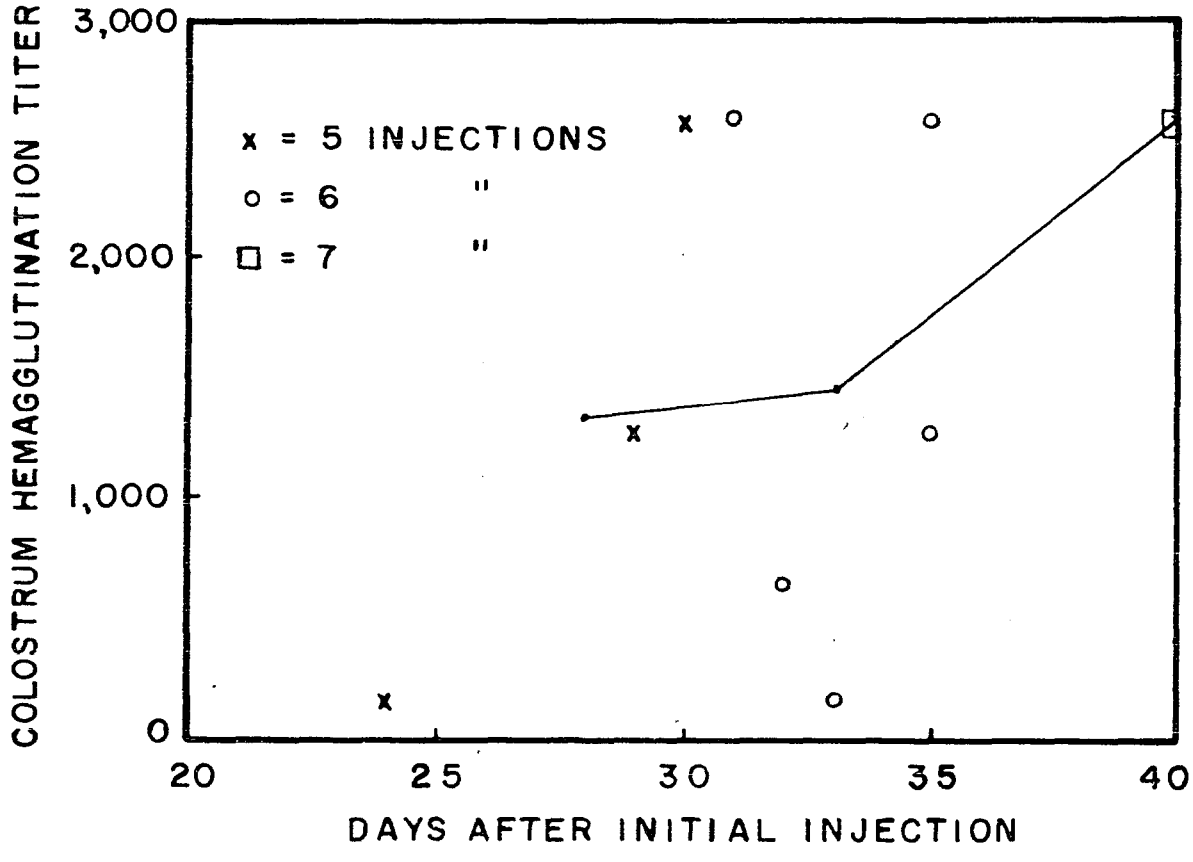


Figure 7. Experiment 778 (Group I). The reciprocal colostrum hemagglutination titers of sows challenged with E. coli antigen versus the number of days after the initial injection of antigen. The points on the line represent the average titer and average number of days after the initial injection for the sows given a particular number of injections.

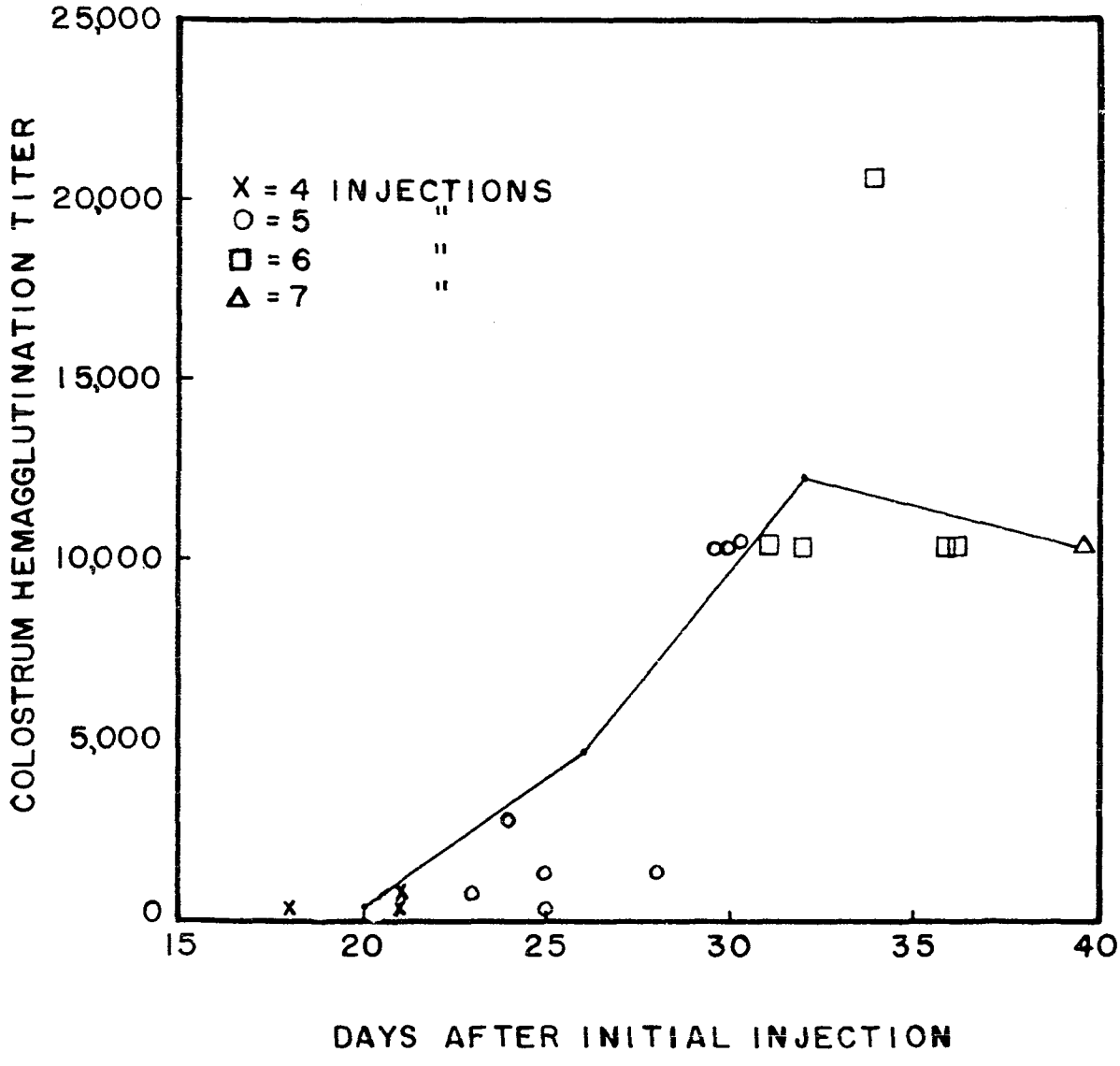


Figure 8. Experiment 778 (Group II). The reciprocal colostrum hemagglutination titers of sows challenged with E. coli antigen versus the number of days after the initial injection of antigen. The points on the line represent the average titer and average number of days after the initial injection for the sows given a particular number of injections.

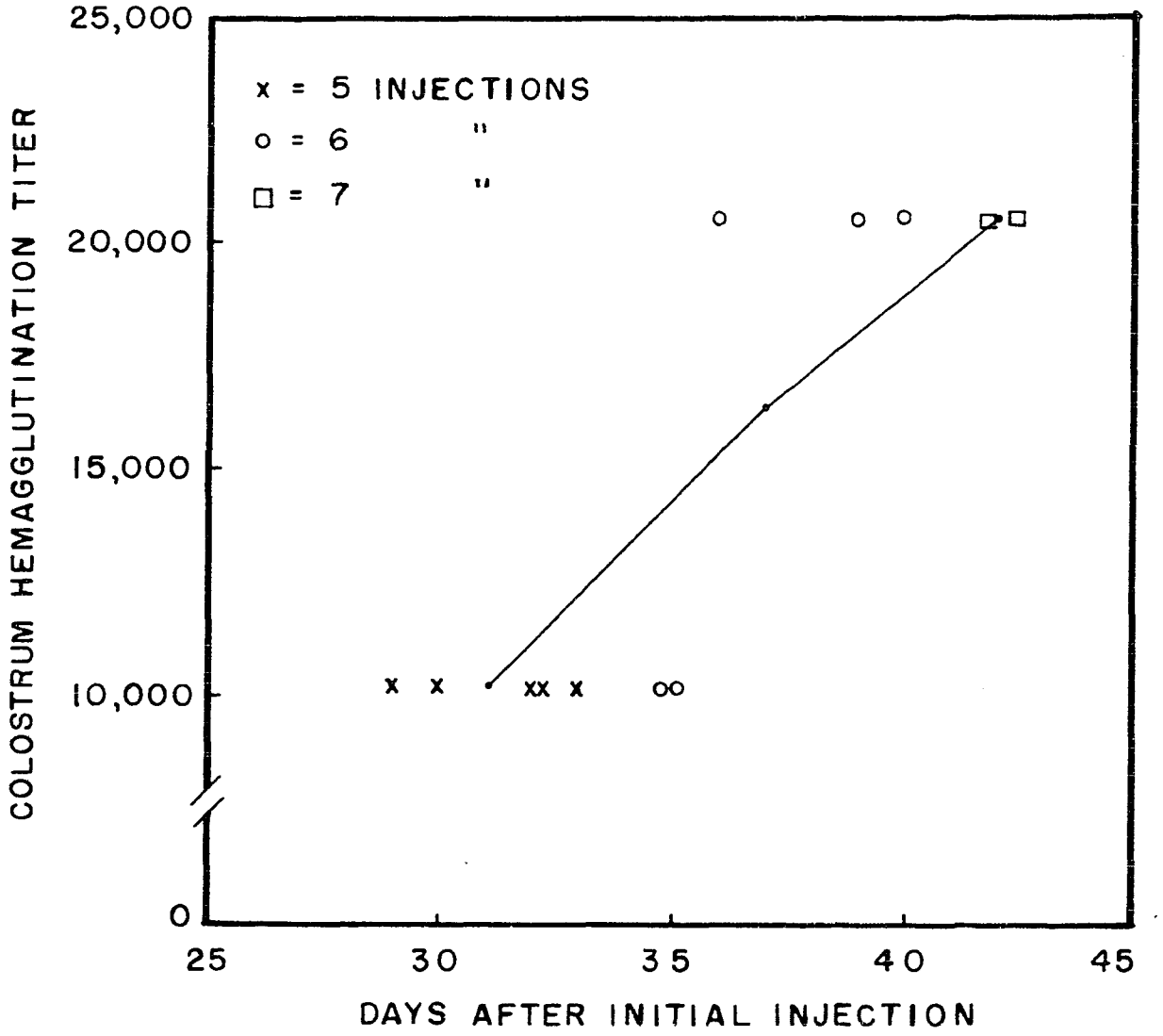


Figure 9. Experiment 778 (Group III). The reciprocal colostrum hemagglutination titers of sows challenged with E. coli antigen versus the number of days after the initial injection of antigen. The points on the line represent the average titer and average number of days after the initial injection for the sows given a particular number of injections.